EVOLUTIONARY GENETIC ANALYSES OF THE FOREST TREE GENUS *POPULUS*

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ABSTRACT

Evolutionary Genetic Analyses of the Forest Tree Genus *Populus*

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Populus, a genus of deciduous trees distributed throughout the northern-hemisphere comprising aspens, poplars and cottonwoods, has recently become a model for forest trees and subject of many interdisciplinary studies in higher plants. However, due to a high level of morphological diversity, extensive inter-species hybridization, and a low level of DNA sequence variation among species in this group, the taxonomy and phylogeny of the genus still remains controversial and incongruent phylogenetic relationships among poplar species have challenged evolutionary biologists for more than a decade.

Nuclear genomic data based upon inter simple sequence repeat (ISSR) as well as six nuclear genome based Sequence Characterized Inter Simple Sequence Repeat (SCISSR) data were used to assess genetic relationships among species of the genus *Populus*. The direction of natural hybridization between two sympatric *Populus* species in North America (*P. deltoides* and *P. balsamifera*) was also investigated using species-specific single nucleotide polymorphism (SNP) markers in both the nuclear and chloroplast genomes.

Overall, the genetic relatedness estimates based on nuclear ISSR data were congruent with phylogenetic trees based on other molecular (RFLP and DNA sequence) and morphological data, but provided better resolution in assessing the genetic relatedness among closely related taxa, and provided genetic evidence for previously suspected introgressions. The resulting SCISSR based tree was also mainly congruent with the genetic relatedness estimates based on other molecular (RFLP, ISSR, and DNA sequence) and morphological data, but showed better resolution of the internal nodes. This result would suggest the need for larger sequence data sets for precise reconstruction of phylogenetic relationships among closely related species of *Populus*.

Although hybridization is generally considered to be symmetrical, with both hybridizing species being equally likely to be the male or female parent, several studies have demonstrated the presence of asymmetrical hybridization and introgression from one species to the other. All natural hybrid individuals, identified based on morphological traits, had nuclear alleles corresponding to both parental species, while the chloroplast genotypes showed similarity to *P. deltoides*, indicating asymmetrical hybridization with *P. deltoides* as the maternal and *P. balsamifera* as the paternal donor species. This observed asymmetrical hybridization may be attributable to cytonuclear interactions.

DEDICATION

The poplars are felled, farewell to the shade

And the whispering sound of the cool colonnade:

The winds play no longer and sing in the leaves,

Nor Ouse on his bosom their image receives.

From The Poplar Field by William Cowper (1731-1800)

To my parents Razi Hamzeh and Shamsi Majlessi

for their endless love and support

To my Husband Reza A-Sadeghi

for his emotional and intellectual support, encouragement and assistance

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GENERAL INTRODUCTION

The genus *Populus* L. is a member of the Salicaceae, which together with the Flacourtiaceae and 29 other families, have been placed under the Malpighiales in the recent cladistic analysis of the angiosperms (The Angiosperm Phylogeny Group 1998). It is a genus of deciduous trees comprising aspens, poplars, and cottonwoods, having a wide natural distribution in the Northern Hemisphere and a small representation in tropical Africa.

Despite many recent advances in the biology of *Populus* (i.e. growth and development, well characterized molecular physiology, genomic, ... etc.), due to its status as a model forest tree, the evolution of the genus has yet to be thoroughly resolved.

An essential component of evolution is natural variation among individuals. Therefore, in order to gain insight into the evolution of a group of taxa, it is important to understand the evolutionary interrelationships among them, trying to interpret the way in which they have diversified and changed over time, how this variation arises and is distributed geographically, and the processes that create distinct units of variation, especially the formation of species.

Phylogeny plays a major role in representing the relationships among groups of biological entities, such as genes and species. A reconstructed phylogeny helps guide interpretation of the evolution of organismal characteristics, providing hypotheses about the lineages in which traits arose and under what circumstances, thus playing a vital role in studies of adaptation and evolutionary constraints (Felsenstein 1985; Maddison 1990; Martins 1995; Liberles et al. 2001; Merritt and Quattro 2001). It also helps explain

patterns and dynamics of speciation, and to some extent, extinction when fossil data are available (Futuyma 1998; Carroll et al. 2001). Phylogenetic systematics is therefore, no less than understanding the history and pattern of relationships among taxa elucidating new theories for the mechanisms of evolution.

The evolutionary history of some taxa of plants may not always be a linear tree. Rather it may be a network, in which there have been reticulate evolutionary events, especially hybrid speciation (Stebbins 1950; Grant 1981; Arnold 1997; Otto and Whitton 2000). Although the evolutionary importance of natural hybridization has always been a contentious issue (Burke and Arnold 2001), botanists as early as the 1930s have accepted natural hybridization as a persistent and important process in plant evolution (Anderson and Hubritch 1938; Anderson 1949; Heiser 1949; Grant 1953; Anderson and Stebbins 1954). This perspective, that has gained support over the past decades, considers hybridization to be a relatively widespread and potentially creative evolutionary process through the production of novel adaptations and new hybrid species (Anderson 1949; Randolph 1966; Knobloch 1971; Harrison 1990; Arnold 1993; Wang et al. 1997). Introgression (the transfer of genetic materials from one species into another via hybridization) has been documented in a wide variety of both plant and animal taxa (Arnold 1992; Rieseberg and Wendel 1993) and there is evidence that it may serve as a source of adaptive genetic variation or adaptive evolution (Lewontin and Birch 1966; Grant and Grant 1994; Grant and Grant 1996; Wang et al. 1997). When hybridization has been directly implicated in the evolutionary process, it has traditionally been for the role it may play in finalizing speciation through reinforcement (Dobzhansky 1940; Mayr 1963; Howard 1993). There are a number of well-documented cases of homoploid hybrid

speciation, suggesting that natural hybridization may play an important role in evolutionary diversification or divergent evolution (Rieseberg 1997). Therefore, studying the pattern of natural hybridization and gene flow between the populations of two distinct, but still reproductively compatible species is crucial to understand the evolutionary history of the interbreeding species or group of taxa.

Species of the genus *Populus*, collectively known as poplars, are an important group of forest trees in North America with a wide distribution pattern from subtropical to boreal forest. The high level of morphological diversity, extensive inter-species hybridization, and low level of DNA sequence variation among species in this group have impeded the progress of taxonomic and phylogenetic studies.

The number of nominal *Populus* species currently described in the literature ranges from 22 to 85 and hundreds of hybrids, varieties, and cultivars exist (Dickmann and Stuart 1983; Eckenwalder 1977a, b, 1996). Discrepancies in the number of species can be attributed to the extensive inter-species hybridization and to the difficulties involved in delineating species boundaries due to morphological diversity and phenotypic plasticity. Eckenwalder (1996) classified the genus *Populus* into 29 species grouped into six sections (*Abaso*, *Aigeiros*, *Leucoides*, *Populus*, *Tacamahaca*, *Turanga*). These are considered to be natural divisions that are generally delineated by major hybridization barriers (Zsuffa 1975; Eckenwalder 1996). However, the phylogenetic affinities between sections and the placement of several species within these sections remain controversial. A cpDNA RFLP analysis of *P. nigra* showed similarity to species of the section *Populus*, but RFLP patterns of nuclear *rDNA* showed affinity to species of the section *Aigeiros* (Smith and Sytsma 1990). Allozyme and pollen competition studies suggested a closer

evolutionary relationship between *P. deltoides* (section *Aigeiros*) and *P. maximowiczii* (section *Tacamahaca*) than between *P. deltoides* and *P. nigra* of the section *Aigeiros* (Rajora 1989; Rajora and Zuffa 1990). Recent phylogenetic analyses of the family *Salicaceae* using DNA sequence data from chloroplast *rbcL* (Azuma et al. 2000) and *ITS* of nuclear rDNA (Leskinen and Alstrome-Rapaport 1999) suggest that *Populus* is a monophyletic group, sister to *Salix*. Although a phylogenetic analysis of poplars using 76 morphological traits of buds, leaves, inflorescences, flowers and fruits supported the monophyly of all sections except for *Tacamahaca*, which resolved into two paraphyletic groups (Eckenwalder 1996), the relationships between sections, as well as relationships among taxa within sections, were only partially resolved. Phylogenetic analysis based on RFLP data suggested that the section *Tacamahaca* is polyphyletic and that the section *Populus* is the terminal clade (Smith 1988). On the other hand, a phylogenetic tree based on DNA sequences from the *ITS* region of the nuclear *rDNA* of four species of *Populus* showed an opposite trend with *P. alba* of the section *Populus* as basal followed by *P. lasiocarpa* of the section *Leucoides* and species of the sections *Aigeiros* and *Tacamahaca* as the terminal clade (Leskinen and Alstrome-Rapaport 1999). Mitochondrial and chloroplast restriction site analysis of four *Populus* species suggested a polyphyletic relationship for species in the section *Aigeiros* (Barrett et al. 1993; Rajora and Dancik 1995). A microsatellite based analysis (Rahman and Rajora 2002) revealed a close genetic relatedness between *P. deltoides* (section *Aigeiros*) and *P. trichocarpa* (section *Tacamahaca*) as well as between *P. balsamifera* (section *Tacamahaca*) and *P. nigra* (section *Aigeiros*), suggesting a polyphyly of the sections *Tacamahaca* and *Aigeiros*. Phylogenetic analyses of the genus *Populus* based on three non-coding regions of cpDNA

trnT-trnF (intron of *trnL*, and intergenic regions of *trnT-trnL* and *trnL-trnF*) showed paraphyly for the section *Populus* (or monophyly with the exception of *P. nigra*) and *Aigeiros* and polyphyly in section *Tacamahaca* and, based on *ITS1* and *ITS2* of nuclear rDNA, monophyly for sections *Populus* and *Aigeiros* and paraphyly in sections *Tacamahaca* (Hamzeh and Dayanandan 2004). Based on chloroplast DNA sequence data, *P. nigra* showed a close affinity to species of the section *Populus*, whereas nuclear DNA sequence data suggested a close relationship between *P. nigra* and species of the section *Aigeiros*, suggesting a possible hybrid origin for *P. nigra*. Similarly, the chloroplast DNA sequences of *P. tristis* and *P. szechuanica* were similar to that of the species of the section *Aigeiros*, while the nuclear sequences revealed a close affinity to species of the section *Tacamahaca,* suggesting a hybrid origin for these two Asiatic balsam poplars. However, due to low level of nucleotide sequence variation between species, the phylogenetic relationships among species within sections remained poorly resolved (Hamzeh and Dayanandan 2004).

Thus, the observed low level of nucleotide sequence variability suggests that the genus *Populus* comprises a group of closely related species, and emphasises the need for a highly variable genetic marker to resolve the phylogenetic relationships among these species.

Extensive interspecific hybridization even between species from different sections (Barnes 1961; Brayshaw 1965; Eckenwalder 1982, 1996; Barnes and Pregitzer 1985; Stettler et al. 1996 a; Whitham et al. 1996) is a well known characteristic of member of the genus.

In North America, several species of the genus *Populus*, particularly species of the sections *Tacamahaca* Spach. and *Aigeiros* Duby, are broadly sympatric (Eckenwalder 1984 a, b), and known to hybridize extensively (Brayshaw 1965; Ronald et al. 1973 a, b; Eckenwalder 1984 a, b; Rood et al. 1986; Greenaway et al. 1991; Floate 2004). Multivariate analysis of leaf morphology of *Populus* species showed continuous variation of characters and suggested hybridization between *P. deltoides*, *P. balsamifera*, and *P. angustifolia* (Rood et al. 1986). These observations were further confirmed with gas chromatography-mass spectrometry data (Greenaway et al. 1991). Comprehensive analyses of leaf morphological characters of *P. balsamifera*, *P. angustifolia* and *P. deltoides* in the hybrid zones in the riparian forests in southern Alberta suggested bidirectional introgression between *P. angustifolia* and *P. balsamifera* (F1 could backcross to both parental species), and unidirectional introgression between *P. balsamifera* and *P. deltoides* (F1 would only backcross to *P. balsamifera*; Floate 2004). Molecular data (RFLP) has also revealed a similar unidirectional introgression between *P. fremontii* and *P. angustifolia* in the Weber River drainage system in northern Utah (F1 would only backcross to *P. angustifolia*; Keim et al. 1989; Martinsen et al. 2001).

Although the hybrid nature and directionality of the hybridization has been successfully determined using morphological and molecular data, the maternal and paternal parental sources of these natural hybrids remain unknown.

With regard to this prologue, there are two major objectives in my dissertation:

1) Resolving the phylogenetic relationships among the closely related species of the genus *Populus* using :

a) Inter Simple Sequence Repeat (ISSR) data.

b) Sequence Characterized Inter Simple Sequence Repeat (SCISSR) data.

2) Characterization of species-specific nuclear and chloroplast SNPs markers for two native eastern North American poplars, *P. deltoides* and *P. balsamifera*, and detecting the direction of natural hybridization between these species.

These objectives will be addressd in three following chapters:

Chapter 1; Genetic relationships among species of *Populus* (Salicaceae) based on nuclear genomic data.

Chapter 2; Phylogeny of the genus *Populus,* based on novel nuclear genomic markers, Sequence Characterized Inter Simple Sequence Repeats (SCISSRs).

Chapter 3; Asymmetrical natural hybridization between *Populus deltoides* and *P. balsamifera* (Salicaceae)

CHAPTER 1; GENETIC RELATIONSHIPS AMONG SPECIES OF *POPULUS* (SALICACEAE) BASED ON NUCLEAR GENOMIC DATA

ABSTRACT

The genus *Populus* comprises some of the most commercially exploited, pioneer forest trees distributed throughout the northern-hemisphere. The high level of morphological diversity, extensive inter-species hybridization, and low level of DNA sequence variation among species in this group have impeded the progress of taxonomic and phylogenetic studies. We used nuclear genomic data based upon inter simple sequence repeat (ISSR) variability, a highly variable class of molecular markers, to determine the genetic relationships among species of the genus *Populus*. Species of the section *Populus* (*Leuce*) clustered together suggesting monophyly of the section *Populus*. The Eurasian members of section *Populus* (*P. alba*, *P. davidiana*, and *P. tremula*) showed closer genetic relationships to each other than to two North American aspens (*P. tremuloides* and *P. grandidentata*) from the same section. In contrast to previous phylogenetic studies, *P. nigra* showed a close genetic relationship to species of the section *Tacamahaca*. This relationship is in agreement with various phenotypic traits, interfertility and the chemistry of bud exudates and serves as evidence for introgression between *P. nigra* and species of the section *Tacamahaca*. Overall, the genetic relatedness estimates based on nuclear ISSR data were congruent with phylogenetic trees based on other molecular (RFLP and DNA sequence) and morphological data. They did, however, provide better resolution for assessing the genetic relatedness among closely related taxa,

and provided genetic evidence for previously suspected introgressions.

Key words: *Populus*; Salicaceae; ISSR markers; Phylogeny; Introgression.

1. 1 INTRODUCTION

The members of the genus *Populus* L. (Salicaceae), commonly known as aspen, cottonwood and poplar, are some of the most widely distributed forest trees in the northern hemisphere. They play an important ecological role as post-fire regenerating pioneer species in boreal forests, and are dominant species in the riparian forests that serve as wildlife habitats and watersheds (Braatne et al. 1992). Because of many inherent features, including fast growth rates, profuse vegetative propagation, adaptability to various ecological conditions, and being a valuable source of wood, the species of *Populus* have become one of the most commercially exploited groups of forest trees in North America and Europe.

Despite many recent advances in understanding the biology of *Populus*, the taxonomy and phylogeny of the genus remains poorly understood. The number of *Populus* species currently recognised in the literature ranges from 22 to 85 and hundreds of hybrids, varieties, and cultivars exist (Eckenwalder 1977a, b, 1996; Dickmann and Stuart 1983). Discrepancies in the number of species can be attributable to the extensive inter-species hybridization and to difficulties involved in delineating species boundaries, due to morphological diversity and phenotypic plasticity.

Eckenwalder (1996) classified the genus *Populus* into 29 species in six sections. These are considered to be natural divisions that are generally delineated by major hybridization barriers (Zsuffa 1975; Eckenwalder 1996). The section *Abaso* Ecken. comprises a single species, *P. mexicana* Ecken., that is confined to Mexico. The section *Turanga* Bunge contains three species and is geographically restricted to Central and

West Asia and North Africa. The section *Leucoides* Spach. also shows a limited geographic distribution with two species in China and the remaining species, *P. heterophylla* L., in the Southeast United States. The section *Aigeiros* Duby contains three species that are widely distributed in Europe and North America. The remaining two widespread sections, *Tacamahaca* Spach. and *Populus* (*Leuce* Duby) are relatively large, and include nine and ten species respectively.

The phylogenetic affinities between sections and the placement of several species within these sections remain controversial. The cpDNA RFLP analysis of *P. nigra* L. showed similarity with species of the section *Populus*, but RFLP patterns of nuclear rDNA showed affinity to species of the section *Aigeiros* (Smith and Sytsma 1990). Recent phylogenetic analyses of the family Salicaceae, using DNA sequence data from chloroplast *rbcL* (Azuma et al. 2000) and ITS of nuclear rDNA (Leskinen and Alstrome-Rapaport 1999), suggest that *Populus* is a monophyletic group sister to *Salix* L. A phylogenetic analysis of poplars, using 76 morphological traits of buds, leaves, inflorescences, flowers and fruits supported the monophyly of all sections except for *Tacamahaca*, which resolved into two paraphyletic groups (Eckenwalder 1996). However, the relationships among sections as well as relationships among taxa within sections were only partially resolved.

Phylogenetic analysis based on RFLP data suggested that section *Tacamahaca* is polyphyletic and that the section *Populus* is considered as an advance clade occupying a terminal position on the phylogenetic tree (Smith and Sytsma 1990). On the other hand, a phylogenetic tree based on DNA sequences from the ITS region of nuclear rDNA of four species of *Populus* showed an opposite trend with *P. alba* L. of section *Populus* as

basal followed by *P. lasiocarpa* Oliv. of section *Leucoides* and species of sections *Aigeiros* and *Tacamahaca* as the terminal clade (Leskinen and Alstrome-Rapaport 1999).

Phylogenetic analyses of the genus *Populus,* based on three non-coding regions of cpDNA *trnT-trnF* (intron of *trnL*, and intergenic regions of *trnT-trnL* and *trnL-trnF*) and ITS1 and ITS2 of nuclear rDNA (Hamzeh and Dayanandan 2004), showed monophyly for the section *Populus* and polyphyly in the sections *Tacamahaca* and *Aigeiros*. Based on chloroplast DNA sequence data, *P. nigra* showed a close affinity to species of the section *Populus*, whereas nuclear DNA sequence data suggested a close relationship between *P. nigra* and species of the section *Aigeiros*, suggesting a possible hybrid origin for *P. nigra*. Similarly, chloroplast DNA sequences of *P. tristis* Fisch. and *P. szechuanica* Schneid. were similar to species of section *Aigeiros*, while the nuclear sequences revealed a close affinity to species in the section *Tacamahaca*, suggesting a hybrid origin for these two Asiatic balsam poplars. However, due to a low level of nucleotide sequence variation between species, the phylogenetic relationships among species within sections remained poorly resolved (Hamzeh and Dayanandan 2004).

Thus, the observed low level of nucleotide sequence variability suggests that the genus *Populus* is a group of closely related species, and emphasises the need for a highly variable genetic marker to resolve the genetic relationships among these species. In general, one of the main challenges in elucidating the genetic relationships among closely related groups of taxa is finding genetic markers with high interspecific polymorphism and little or no intra-specific variation that can be used to resolve evolutionary relationships among species. In the present study, we have used nuclear genome based ISSR (inter simple sequence repeat) markers, a highly variable, low cost and highly

reproducible class of molecular markers to determine the genetic relationships among species of *Populus*.

The ISSR markers consist of PCR amplification products of DNA sequences flanked by two inverted microsatellites (Zietkiewicz et al. 1994). These markers are generated from single-primer PCR reactions where the primer is designed from di-or tri-nucleotide repeat motifs with a 3' anchoring sequence of one to three nucleotides (Gupta et al. 1994; Zietkiewicz et al. 1994). Since di- and tri-nucleotide SSRs are not found in vascular plant mitochondrial or chloroplast genomes (Wolfe et al. 1998), ISSR primers are used in this study to represent nuclear molecular markers. The ISSRs show excellent reproducibility and high levels of polymorphism (Kojima et al. 1998; Bornet and Branchard 2001) and are being used in numerous applications, ranging from population to interspecific genetic studies (Wolfe et al. 1998).

The present study is focused on species of the large (*Tacamahaca* and *Populus*), and small but geographically widespread (*Aigeiros*) sections in which evolutionary genetic relationships among species remained unresolved in previous studies (Hamzeh and Dayanandan 2004). These included several ecologically and economically important species, some of which are widely used in plantations in North America and Europe. Tissue samples of *Populus* species with restricted geographical ranges were not available for the present study

1. 2 MATERIALS AND METHODS

1. 2. 1 Taxon sampling, DNA extraction, and PCR amplification.

Taxon sampling was based on the classification of the genus *Populus* proposed by Eckenwalder (1977a, b, 1996) and Dickmann and Stuart (1983). Fresh leaves were obtained from 21 taxa including 17 species representing three sections of the genus *Populus* (sections *Tacamahaca*, *Aigeiros*, and *Populus*), and two varieties of *P. deltoides* Bartr. ex Marshall. Depending upon availability, we selected up to three individuals per species to discern the effect of intraspecific ISSR variability. Two species of *Salix* (*S. integra* Thunb. and *S. nigra* Marshall) were used as outgroups. The list of species used in this study and the accession numbers are given in Table 1. 1 The total genomic DNA was extracted from frozen leaf tissue using the methods of Doyle and Doyle (1987) and Dayanandan et al.(1997). Seven ISSR primers were used in single-primer PCR reactions (Table 1. 2). Three primers were specifically designed for the present study, and the remaining four primers were from Wolfe et al. (1998). These primers were synthesized by Operon Technologies (Alameda, California, USA). The amplification reactions contained 230 μM dNTP, 2.5 mM $MgCl_2$, 0.4 μM of a given ISSR primer, 1 unit of Taq DNA polymerase, and 2.5μl buffer (0.2M Tris pH.9.5; 0.25 M KCl; 1mg/ml BSA, 5μl /ml tween 20) in a total volume of 25 μl. PCR amplification was performed in a Mastercycler gradient thermal cycler (Eppendorf, Westbury, New York, USA) with the touchdown thermal cycling protocol starting at 5°C above optimum annealing temperature and a 1°C /cycle temperature decrease down to optimum annealing temperature. The remaining thermal cycling protocol was set to 94°C for 60 s, optimum

annealing temperature for 30 s, and extension at 72°C for 60 s for a total of 38 cycles. The optimum annealing temperature of each ISSR primer is given in Table 1. 2.

1. 2. 2 Data collection and phylogenetic analysis.

Ten µl of each PCR product were electrophoresed on a 1% agarose gel with ethidium bromide (0.33µg/ml) at 3.5 v/cm for 90 minutes. ISSR bands were visualized and documented digitally using a GeneSnap 4.00–Gene Genius Bio Imaging System (Syngene, Frederick, Maryland, USA). The digital image files were analyzed using Gene Tools software (Syngene) and fragment sizes were estimated based on a GeneRuler 1kb DNA Ladder (Fermentas, Burlington, Ontario, Canada). An example of the analysis procedure is given in Appendix 1. 1. The DNA fragments on each agarose gel were scored as binary characters (1 = band present; 0 = band absent) and analyzed using PAUP* v: 4.0 beta 8 (Swofford 2001). The genetic relationships among samples were elucidated using the neighbor-joining (NJ) algorithm as implemented in PAUP with the mean character value as a distance measure. The robustness of the branches was tested via bootstrap analysis of 1000 replicates using PAUP.

1. 3 RESULTS

The ISSR DNA bands scored resulted in a binary data matrix consisting of 79 characters and 40 taxa. The total number of bands scored as present per taxon ranged from 8 to 31 and the average number of bands scored as present per primer ranged from 9.5 to 14.4. The size of the ISSR bands ranged from 200 to 3400 bp (Table 1. 2). Genotyping was repeated twice and only consistently reproducible bands were scored.

In the dendrogram based on all seven ISSR primers using the neighbor-joining method (Fig. 1. 1), all *Populus* species sampled formed a monophyletic group and multiple individuals (with the exception of *P. trichocarpa* Torr. & Gray ex Hook) of each species clustered together. The section *Populus* was monophyletic, while the section *Tacamahaca* was paraphyletic. *Populus tristis* of section *Tacamahaca* grouped within the clade comprising species of section *Aigeiros*. In section *Populus*, the Eurasian members of the section namely *P. alba*, *P. davidiana* Schneid, and *P. tremula* L. showed closer relationships to each other than to the two North American aspens (*P. tremuloides* Michx. and *P. grandidentata* Michx.) of section *Populus*. *Populus nigra* clustered with a group consisting of *P. balsamifera* L., *P. angustifolia* James, and *P. szechuanica* of section *Tacamahaca*. *Populus szechuanica*, one of the Asiatic balsam poplars, was placed in the group of North American balsam poplars (*P. balsamifera* and *P. angustifolia*) in section *Tacamahaca*. North American cottonwoods of section *Aigeiros* clustured with species of section *Tacamahaca* and showed a close genetic relatedness to *P. tristis* and *P. trichocarpa*.

1. 4 DISCUSSION

The resulting dendrogram (Fig.1. 1) showed monophyly of species of genus *Populus*, monophyly of species in section *Populus*, polyphyletic relationship among species of section *Aigeiros* and paraphyly of section *Tacamahaca*. Although the bootstrap support for the monophyly of the genus *Populus* based on ISSR data is not strong, previous studies based on cpDNA and rDNA data strongly supported the monophyly of the genus (Hamzeh and Dayanandan 2004). Since the objective of the present study was to use the hypervariable ISSR genomic regions to resolve the genetic relationships of the taxa in the terminal branches, low bootstrap support at basal nodes based upon ISSR data is not unexpected. Thus, the congruence between different data sets is invaluable in assessing the overall evolutionary relationships of the genus *Populus*. The monophyly of the section *Populus* is in agreement with other nuclear DNA based phylogenetic studies, but incongruent with the cpDNA sequence-based phylogeny in which *P. nigra*, a taxon considered to be a member of section *Aigeiros*, clustered with species in section *Populus* (Smith and Sytsma 1990; Hamzeh and Dayanandan 2004). The evolutionary genetic relationships between sections *Aigeiros* and *Tacamahaca* have long been controversial. Species of these two sections are known to be freely inter-fertile (Zsuffa 1975; Eckenwalder 1984a). The close genetic relationship between *P. deltoides* and *P. maximowiczii* A. Henry is supported by isozyme data (Rajora and Zuffa 1990). The ISSR-based dendrogram revealed a paraphyletic relationship among species of the section *Tacamahaca*. This is consistent with chloroplast DNA and nuclear rDNA

sequence data (Hamzeh and Dayanandan 2004), microsatellite data (Rahman and Rajora 2002), and morphology-based phylogenetic analyses (Eckenwalder 1996).

The dendrogram based on ISSR data suggests a close affinity between *P. deltoides* (section *Aigeiros*) and *P. tristis* (section *Tacamahaca*). The cpDNA- and nuclear rDNA-based phylogeny (Hamzeh and Dayanandan 2004) suggested that *P. tristis* (Himalayan balsam poplar) may have been derived from an ancient hybridization event with an ancestor of species of the section *Aigeiros* as the maternal (cpDNA) donor and, probably the immediate ancestor of the lineage comprising species of section *Tacamahaca* (North American *P. angustifolia, P. balsamifera, P. trichocarpa*, and Chinese *P. cathayana* Rehder and *P. szechuanica*) as a paternal donor. Based on chloroplast RFLP data and a preliminary analysis of nuclear rDNA RFLP, Smith and Sytsma (1990) concluded that *P. tristis* is an introgressant or hybrid of the *P. nigra* (cpDNA) lineage and the Asian members of the section *Tacamahaca.* The ISSR-based dendrogram of the present study links *P. tristis* with the North American cottonwood lineage (*P. deltoides*, *P. sargentii* Dode, *P. angulata* Ait., and *P. fremontii* S. Watson). It also shows an affinity to *P. trichocarpa* (black cottonwood), a North American member of the section *Tacamahaca*, which shows a closer relatedness to species of the section *Aigeiros* than to other species of the section *Tacamahaca.* Since the ISSR primers used in this study represent nuclear molecular markers, the data generated through ISSR analysis are representative of both maternal and paternal lineages and valuable for detecting introgression and hybrid lineages. Therefore, the evolutionary genetic analysis based on various methods including ISSR data, along with the high morphological similarity of *P. tristis* to North American *P. balsamifera* and the existence of a hybrid

clone *P. tristis* x *P. balsamifera* (Dickmann and Stuart 1983) strengthen the hypothesis that *P. tristis* is a taxon of hybrid origin. Since our sample size of *P. tristis* is limited, further studies involving multiple samples are needed to confirm these findings.

The European black poplar, *P. nigra* is considered to be one of the most controversial species in the genus *Populus* with affinities to poplars (section *Populus*) and cottonwoods (section *Aigeiros*). The RFLP analysis of chloroplast DNA (Smith and Sytsma 1990), and chloroplast DNA sequence data (Hamzeh and Dayanandan 2004) indicated a close affinity between *P. nigra* and species of the section *Populus*, whereas RFLP (Smith and Sytsma 1990) and nuclear rDNA sequence data (Hamzeh and Dayanandan 2004), and morphology (Eckenwalder 1996) indicated a close affinity between *P. nigra* and species of section *Aigeiros*. In the ISSR based dendrogram, *P. nigra* is closely related to the group comprising *P. balsamifera*, *P. angustifolia*, and *P. szechuanica* (section *Tacamahaca*). This relationship is further supported by interfertility and bud exudate data. Based on bud exudate data, Eckenwalder (1996) suggested a close affinity between *P. nigra* and species of section *Tacamahaca*. Isozyme (Rajora and Zuffa 1990) and pollen competition data (Rajora 1989) also suggest a close affinity between species of the sections *Tacamahaca* and *Aigeiros*. In addition, Dickmann et al. (2001) pointed out that crossing between *P. nigra* and species of the section *Populus* is problematic. However, *P. nigra* can hybridize with *P. deltoides* and *P. fremontii* (section *Aigeiros*) as well as with *P. angustifolia* and *P. trichocarpa* of the section *Tacamahaca* (Eckenwalder 1996). Thus, the placement of *P. nigra* in sections *Populus*, *Aigeiros*, or *Tacamahaca* remains controversial.

It is generally assumed that the degree of relative inter-fertility is a reflection of genetic relatedness (Stettler et al. 1996 a; Dickmann et al. 2001) and, therefore, can form a basis for inferring taxonomic affinities. Interspecific hybridization within the genus *Populus* is generally limited to intrasectional crosses, or intersectional crosses between species of *Aigeiros* and *Tacamahaca* (Ronald 1982; Rajora 1989). Species of the section *Populus* are reproductively isolated from species of other sections and exhibit intersectional incompatibility reflecting evolutionary divergence between species of the section *Populus* and those in other sections including *Aigeiros* and *Tacamahaca* (Eckenwalder 1996). Intersectional crosses between *Tacamahaca* and *Aigeiros* are generally compatible in most combinations (Zsuffa 1975) and have given rise to many vigorous clones used in commercial plantations (Dickmann and Stuart 1983).

The ISSR-based dendrogram revealed a close genetic relationship between North American cottonwoods of section *Aigeiros* and *P. trichocarpa*, the North American black cottonwood of section *Tacamahaca*. Species of the sections *Tacamahaca* and *Aigeiros* are sympatric in North America (Little 1971) with overlapping ecological preferences (Fowells 1965). The North American representatives of *Aigeiros* and *Tacamahaca* show similarities in floral traits, and can hardly be distinguished on the basis of floral morphology (Eckenwalder 1977a, 1984b). The ISSR data reveals a close relationship between *P. trichocarpa* and *P. deltoides*. The existence of natural and artificial hybrids between *P. deltoides* and *P. trichocarpa* (Stettler et al. 1996b) further supports their genetic and evolutionary affinities. *P.* x *generosa* Henry, a hybrid between *P. trichocarpa* and *P. deltoides*, is more productive than both parental species, and has

become the basis for hardwood pulp and timber industry in the pacific Northwest U.S. (Heilman and Stettler 1985; Dickmann et al. 2001).

Phylogenetic relationships among North American cottonwoods of section *Aigeiros* showed that *P. fremontii*, *P. sargentii* and *P. angulata* are more closely related to each other than to *P. deltoides*. Although *P. sargentii* (commonly known as plains cottonwood) was originally classified as a variety of *P. deltoides*, it was later considered to be a distinct species (Little 1979). However, this taxon is often classified as *P. deltoides* var. *occidentalis* Rydb or *P. deltoides* ssp *monilifera* (Ait.) Ecken. Similarly, *P. angulata* is classified as *P. deltoides* var. *angulata* Ait. (Dickmann and Stuart 1983; Eckenwalder 1996). ISSR data suggests that *P. sargentii* and *P. angulata* are genetically distinct from *P. deltoides* ssp. *deltoides*.

In conclusion, we have used nuclear genome based ISSR markers to assess genetic relationships among species of the genus *Populus*, a closely related group of species, in which other genetic markers showed limited success in resolving the evolutionary relatedness. The multiple individuals of a given species clustered together, and the resulting dendrogram was congruent with existing phylogenetic trees of poplars based on other molecular, and morphological data. The phylogenetic tree based on seven ISSR markers showed higher resolution than RFLP and DNA sequence based phylogenetic trees. Moreover, due to wide genome coverage, ISSR markers, in conjunction with uniparentally inherited organelle gene sequence data are valuable tools for the detection of introgression and reticulate evolutionary events.

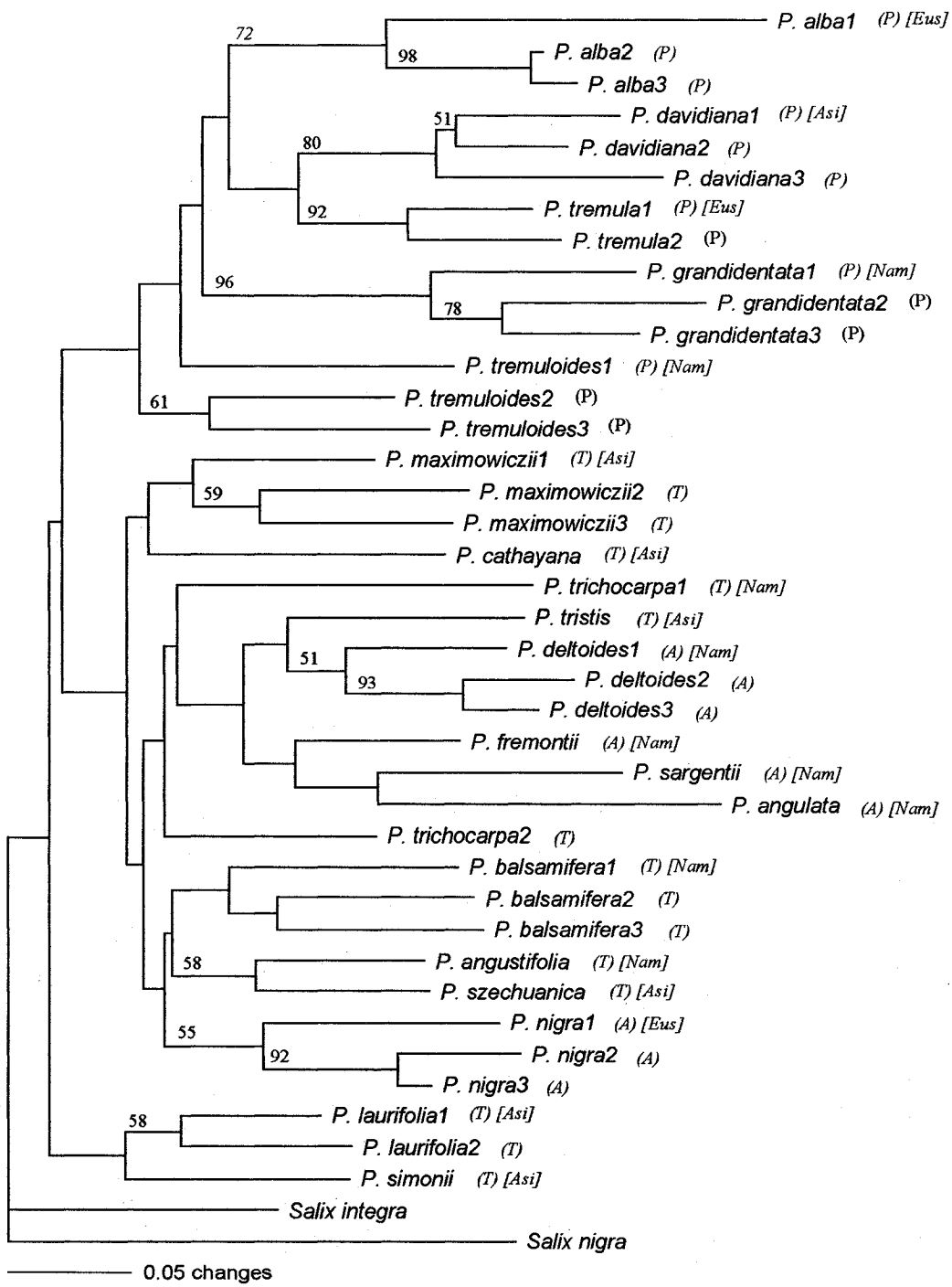
Table 1. 1 Taxa of genus *Populus* L. and outgroups sampled for this study, their common names, voucher numbers, source, and geographic origin (the classification system of *Populus* follows Eckenwalder 1977a, b, 1996, and Dickmann and Stuart, 1983; AP, Alberta Pacific Forest Products Collection; MRNFQ, Le Ministere des Ressources Naturelles et de la Faune du Quebec; MBG, Montreal Botanical Garden; MAM, Morden Research Station and the Arboretum, Manitoba; MAQ, Morgan Arboretum, Quebec).

Taxon	Common name	Voucher number	Source	origin
Sect. *Populus (Leuce* Duby) subsect. Albidae				
P. alba L.	White poplar	MH-002, MH-003, MH-004	AP	Eurasia & N. Africa
Sect. *Populus* (*Leuce* Duby) subsect. Trepidae				
P. davidiana Schneid.	Korean aspen	MH-007, MH-008, MH-009	AP	Asia
P. grandidentata Michx.	Bigtooth aspen	MH-013, MH-014, MH-015	MRNFQ	North America
P. tremuloides Michx.	Trembling aspen	MH-024, MH-025, MH-026	AP	North America
P. tremula L.	European aspen	MH-019, MH-020	AP	Eurasia & N. Africa
Sect. *Tacamahaca* Spach.				
P. angustifolia James	Narrowleaf cottonwood	MH-059	MAM	North America
P. balsamifera L.	Balsam poplar	MH-054, MH-055, MH-056	AP	North America
P. cathayana Rehder		MH-060	MA	Asia
P. laurifolia Ledeb.	Laurel poplar	MH-061, MH-062	MA	Asia
P. maximowiczii A. Henry	Japanese poplar	MH-030, MH-031, MH-032	MRNFQ	Asia
P. simonii Carriere	Simon poplar	MH-065	MBG	Asia
P. szechuanica C. K. Schneid.	Szechuan poplar	MH-066	MBG	Asia
P. trichocarpa Torr. & Gray ex Hook	Black cottonwood	MH-040, MH-040	MRNFQ	North America
P. tristis Fisch.	Himalayan balsam poplar	MH-067	MBG	Asia
Sect. *Aigeiros* Duby				
P. deltoids Bartr. ex Marsh.	Eastern cottonwood	MH-049, MH-050, MH-051	AP	North America
P. deltoids var . *angulata* Ait.	Alamo cottonwood	MH-071	MAM	North America
P. sargentii Dode (*P. deltoides* var. *occidentalis* Rydb.)	Plains cottonwood	MH-070	MAM	North America
P. fremontii S. Watson	Fremont cottonwood	MH-063	MAM	North America
P. nigra L.	Black poplar	MH-043, MH-044, MH-045	MRNFQ	Eurasia
Outgroup taxa				
Salix nigra Marshall		MH-S-001	MAQ	
Salix integra Thunb.		MH-S-002	MAQ	

Table 1. 2 Sequences of ISSR primers, optimum annealing temperature for each ISSR primer, and the number and size range of ISSR bands scored. * Primers from Wolfe et al., (1998).

Primer sequence	Optimum annealing temperature (°C)	Total number of DNA fragments produced by each primer and scored as present	Size of DNA fragments (bp)
(ATC) 10-RC	55	9	500-3400
(AAC) 10-RC	55	14	200-2000
*(CA) 6-RG	45	12	400-1700
*(CT) 8-RG	45	11	400-2700
*(GT) 6-RG	45	10	400-1400
*(CTC) 4-RC	45	13	400-1900
(AGC) 10-RC	60	10	500-2400

Figure 1. 1 Neighbor-joining tree based on distance matrix generated from bands of seven ISSR markers. Numbers listed above branches are bootstrap values >50% based on 1000 replicates. Branches without numbers had bootstrap values <50%. Section affiliations are denoted as A (*Aigeiros*) P (*Populus*), T (*Tacamahaca*). Geographic origins of species indicated (only next to the first individual of a taxon) as Asi (Asia), Nam (North America), Eus (Eurasia).


P. alba1 (P) [Eus]
P. alba2 (P)
P. alba3 (P)
P. davidiana1 (P) [Asi]
P. davidiana2 (P)
P. davidiana3 (P)
P. tremula1 (P) [Eus]
P. tremula2 (P)
P. grandidentata1 (P) [Nam]
P. grandidentata2 (P)
P. grandidentata3 (P)
P. tremuloides1 (P) [Nam]
P. tremuloides2 (P)
P. tremuloides3 (P)
P. maximowiczii1 (T) [Asi]
P. maximowiczii2 (T)
P. maximowiczii3 (T)
P. cathayana (T) [Asi]
P. trichocarpa1 (T) [Nam]
P. tristis (T) [Asi]
P. deltoides1 (A) [Nam]
P. deltoides2 (A)
P. deltoides3 (A)
P. fremontii (A) [Nam]
P. sargentii (A) [Nam]
P. angulata (A) [Nam]
P. trichocarpa2 (T)
P. balsamifera1 (T) [Nam]
P. balsamifera2 (T)
P. balsamifera3 (T)
P. angustifolia (T) [Nam]
P. szechuanica (T) [Asi]
P. nigra1 (A) [Eus]
P. nigra2 (A)
P. nigra3 (A)
P. laurifolia1 (T) [Asi]
P. laurifolia2 (T)
P. simonii (T) [Asi]
Salix integra
Salix nigra
72
98
51
80
92
96
78
61
59
51
93
58
55
92
58
0.05 changes

Appendix 1. 1 Image document data of 1% agarose gel electrophoresis of PCR products using ISSR 3.

SynGene GeneTools - Version 3.00.22 - Serial No. 3984*2001*fishca*mpcs
SynGene Laboratories
This heading can be changed by the user.

Image document data

File	C:\My Documents\Daya\Mona ISSR images\ISSR 3 a.sgd
Created by	GeneGenius
Generated on	Monday, February 10, 2003 01:38:21 PM
ID	1044902301

Black:14 White:241 Gamma:1.00

Properties

User Field 1
User Field 2
User Field 3
User Field 4
User Field 5

Capture	
Digitizer	prymo32
Camera	GeneGenius
Settings	Exposure Time: 0.240 seconds
	Frame accumulation: Off
	(Stop on saturation: On)
	(Stick on saturation: Off)
	Darkroom camera filter: EtBr/UV
	Digitiser offset: 128
	Digitiser gain: 0

User: Mona Created: Monday, December 10, 2007 02:07:19 PM ID : 605492854
Signature : Printed: Monday, December 10, 2007 02:29:36 PM : Page 1

SynGene GeneTools - Version 3.00.22 - Serial No. 3984*2001*fishca*mpcs
SynGene Laboratories
This heading can be changed by the user.

Digitiser range: 128
Facility 1 level: 0
Facility 2 level: 0
Facility data 0: HIGH
Facility data 1: HIGH
Lamp Switch: LOW

Integration parameters

Baseline correction method	LOWEST SLOPE
Baseline correction offset	YES
Color filtration options	NO FILTER
Smoothing	SAVITSKY-GOLAY order 2 width 3
Type	Fluorescence
Image dimensions	780 x 574
Electrophoresis direction	Up
Minimum peak height	3
Minimum peak width (pixels)	7
Minimum peak area	1
Number of tracks	23

Track 1			
Number	Mol. weight	Height	Raw vol.
1(m)	3000.00	188.231	16291.50
2(m)	2500.00	149.582	6236.25
3(m)	2000.00	144.964	12105.74
4(m)	1500.00	110.550	11826.93
5(m)	1000.00	73.483	10268.24
6(m)	750.00	88.633	10023.05
7(m)	500.00	73.653	10904.03
8(m)	250.00	60.716	13257.46

Track 2			
Number	Mol. weight	Height	Raw vol.
1(m)	1275.70	60.583	7252.89
2(m)	845.70	155.677	22023.04
3(m)	606.61	73.629	10031.51

Track 3			
Number	Mol. weight	Height	Raw vol.
1(m)	1329.48	178.945	40656.07
2(m)	866.85	70.271	7143.26
3(m)	595.40	70.869	9254.25

Track 4			
Number	Mol. weight	Height	Raw vol.
1(m)	2508.95	81.149	11972.13
2(m)	1544.49	66.572	10222.49
3(m)	868.06	54.515	3777.04

Track 5			
Number	Mol. weight	Height	Raw vol.
1(m)	1334.47	114.216	16185.66
2(m)	849.54	70.549	8626.97

User: Mona Created: Monday, December 10, 2007 02:07:19 PM ID : 605492854
Signature : Printed: Monday, December 10, 2007 02:29:36 PM : Page 2

Track 6			
Number	Mol. weight	Height	Raw vol.
1(m)	1338.75	174.903	30565.77
2(m)	852.30	55.005	5784.22

Track 7			
Number	Mol. weight	Height	Raw vol.
1(m)	1648.43	64.206	16029.07
2(m)	856.00	81.402	10358.97

Track 8			
Number	Mol. weight	Height	Raw vol.
1(m)	1439.84	47.032	4733.48

Track 9			
Number	Mol. weight	Height	Raw vol.
1(m)	1911.02	118.359	15052.65
2(m)	1451.23	46.394	2483.99

Track 10			
Number	Mol. weight	Height	Raw vol.
1(m)	2789.83	162.652	24842.08
2(m)	1400.37	46.935	4146.55

Track 11			
Number	Mol. weight	Height	Raw vol.
1(m)	2825.82	54.382	5040.15
2(m)	1412.39	66.216	5969.65

Track 12			
Number	Mol. weight	Height	Raw vol.
1(m)	2866.33	43.579	2272.04
2(m)	2267.47	49.426	4179.00
3(m)	1425.87	124.671	14557.11

Track 13			
Number	Mol. weight	Height	Raw vol.
1(m)	1409.01	167.976	33047.15
2(m)	857.39	170.329	18665.00
3(m)	692.72	68.299	6950.48

Track 14			
Number	Mol. weight	Height	Raw vol.
1(m)	2769.38	47.101	3952.52
2(m)	1969.51	53.250	4368.96
3(m)	1681.51	107.631	9331.44
4(m)	845.61	58.262	5826.29

Track 15			
Number	Mol. weight	Height	Raw vol.
1(m)	1989.71	49.785	2557.89
2(m)	1344.32	52.996	8851.63
3(m)	833.56	56.854	7452.81

Track 16			
Number	Mol. weight	Height	Raw vol.
1(m)	2667.46	78.250	10674.21
2(m)	1628.57	117.478	12787.77
3(m)	1192.22	59.057	4953.17
4(m)	839.96	72.432	8660.74

Track 17			
Number	Mol. weight	Height	Raw vol.
1(m)	1639.77	68.450	3434.53
2(m)	1419.71	173.393	19986.28
3(m)	826.31	77.064	9404.94

Track 18			
Number	Mol. weight	Height	Raw vol.
1(m)	2789.16	95.229	8957.76
2(m)	2034.63	167.137	37486.57
3(m)	1366.49	66.155	3409.64
4(m)	830.28	89.799	20684.44

Track 19			
Number	Mol. weight	Height	Raw vol.
1(m)	1936.07	82.927	13802.21
2(m)	1313.67	74.399	11541.34
3(m)	833.27	94.322	12849.11
4(m)	597.59	68.269	7914.62

Track 20			
Number	Mol. weight	Height	Raw vol.
1(m)	1993.60	75.553	8095.88
2(m)	1892.00	109.288	5221.88
3(m)	1288.89	73.000	3601.01
4(m)	1209.06	82.686	6391.61
5(m)	817.25	100.911	11119.24

Track 21			
Number	Mol. weight	Height	Raw vol.
1(m)	2734.52	76.863	5571.13
2(m)	1945.78	129.418	16156.04
3(m)	1346.88	59.206	4065.34
4(m)	836.81	69.905	8976.60
5(m)	587.53	66.746	8611.75

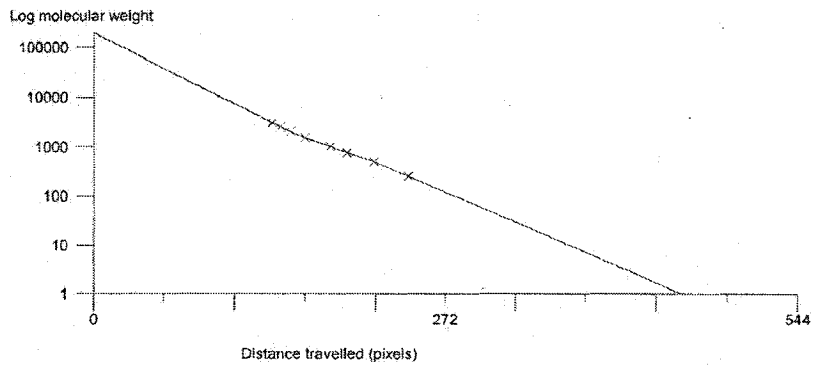
Track 22			
Number	Mol. weight	Height	Raw vol.
1(m)	2114.05	68.265	6346.58
2(m)	1897.47	66.312	5081.70
3(m)	1319.42	63.378	5390.01
4(m)	819.20	67.527	7594.64
5(m)	587.92	56.994	4924.95

Track 23			
Number	Mol. weight	Height	Raw vol.
1(m)	3000.00	190.703	16469.14
2(m)	2500.00	166.590	7783.66
3(m)	2000.00	166.663	12797.27
4(m)	1500.00	138.318	11043.84
5(m)	1000.00	81.435	9027.87
6(m)	750.00	101.664	14575.00
7(m)	500.00	81.124	11779.63
8(m)	250.00	65.656	10593.88

Molecular weight calibration details

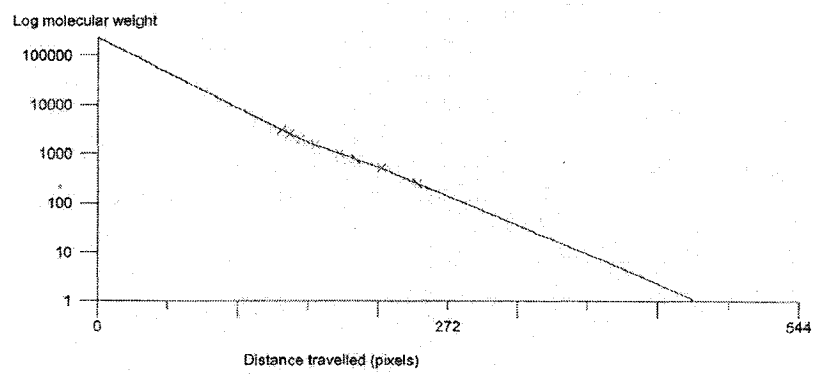
Curve type	Log piecewise linear
Distance measurement	From Rf baseline
Propagation method	Interpolate between standards

Molecular weight standard: Track 1



Track 1	
Peak	Assigned MW.
0	3000.00
1	2500.00
2	2000.00
3	1500.00
4	1000.00
5	750.00
6	500.00
7	250.00

Molecular weight standard: Track 23



Track 23	
Peak	Assigned MW.
0	3000.00
1	2500.00
2	2000.00
3	1500.00
4	1000.00
5	750.00
6	500.00
7	250.00

Quantity calibration details

Curve type	Linear through origin (single standard value)
Calibrate	All tracks to a single curve
Units	

No quantity calibrations are defined !

CHAPTER 2; PHYLOGENY OF THE GENUS *POPULUS* BASED ON THE NOVEL NUCLEAR GENOMIC MARKERS, SEQUENCE CHARACTERIZED INTER SIMPLE SEQUENCE REPEATS (SCISSRS).

ABSTRACT

For many reasons, *Populus* , a genus of deciduous trees which includes aspens, poplars and cottonwoods, has recently become a model for forest trees and has been the subject of many interdisciplinary studies of higher plants. However, the taxonomy and phylogeny of the genus still remains controversial and incongruent phylogenetic relationships among poplar species have challenged evolutionary biologists for more than a decade.

We have used a combined set of six nuclear genome based Sequence Characterized Inter Simple Sequence Repeat (SCISSR) data to assess genetic relationships among species of the genus *Populus*. Based on this analysis all *Populus* species formed a strongly supported monophyletic group consisting of two major well supported clades. One clade included all of the species of section *Populus*, indicating the monophyletic origin of this section. In the second clade one strongly supported group included all of the North American cottonwoods of section while the other group included all of the species of section *Tacamahaca* plus the controversial member of section *Aigeiros*, *P. nigra*. The resulting phylogenetic tree although mainly congruent with the genetic relatedness estimates based on other molecular (RFLP, ISSR, and DNA sequences) and morphological data, showed a better resolution of the internal nodes. This

result would suggest the need for larger sequence data sets for precise reconstruction of phylogenetic relationships among closely related species of *Populus*.

2. 1 INTRODUCTION

The genus *Populus* L. is a member of the family Salicaceae, which, together with 30 other families, has been placed in the order Malpighiales in the recent cladistic analysis of the angiosperms (The Angiosperm Phylogeny Group 1998). *Populus* is a genus of deciduous trees consisting of aspens, poplars and cottonwoods. It has a wide natural distribution in the boreal forest region and in warm temperate to subtropical latitudes across the northern hemisphere. It also has a small representation in the tropics (eg, *P. mexicana* and *P. ilicifolia*; Eckenwalder 1996). For simplicity the term "poplars" will be used hereafter unless a specific subgroup is discussed.

Poplars are dioecious, wind-pollinated trees producing large amounts of pollen and small cotton-tufted seeds, which are dispersed by wind or water in early summer. Moreover, all poplars can reproduce asexually by sprouting from the root collar or sucker shoots arising from horizontal roots (Dickmann et al. 2001).

Poplars because of their rapid growth, abundant natural genetic variation, ease of sexual reproduction and interspecific hybridization, ease of cloning, the small genome that has been completely sequenced, and the ease of transgenesis have recently become model forest trees and the subject of many interdisciplinary studies in higher plants (Bradshaw et al. 2000). However, extensive inter-species hybridization (Barnes and Pregitzer 1985; Eckenwalder 1982, 1984 a, b, c,1996; Whitham et al. 1996) and morphological diversity is the cause of discrepancies in the number of poplar species mentioned in the literature, ranging from 22 to 85 plus hundreds of hybrids, varieties, and cultivars (Dickmann and Stuart 1983; Eckenwalder 1977 a, b, 1996). Moreover, the

generality of species definition is limited due to between species divergence (Hart and Sunday 2007). According to Eckenwalder (1996) the genus can be classified into 29 species grouped into six sections (*Abaso*, *Aigeiros*, *Leucoides*, *Populus*, *Tacamahaca*, and *Turanga*). These are considered to be natural divisions that are generally delineated by major hybridization barriers (Zsuffa 1975; Eckenwalder 1996). However, the phylogenetic origin of the sections and placement of several species within these sections remains controversial.

One of the most persistent challenges to molecular phylogenetic reconstruction is the widespread incongruence between alternative phylogenies obtained using different single-gene/ genomic region data sets. Incongruence occurs at all taxonomic levels, from phylogenies of closely related species (Mason-Gamer and Kellogg 1996; Kopp and True 2002) to relationships between major classes (Giribet et al. 2001; Hwang et al. 2001) or phyla and higher taxonomic group (Baldauf and Palmer 1993; Loytynoja and Milinkovitch 2001; Rokas et al. 2003 a). Both analytical and biological factors may cause incongruence (Wendel and Doyle 1998; Rokas et al. 2003 a). Analytical factors, such as the choice of optimality criteria (Huelsenbeck 1995), limited data availability (Philippe et al 1994; Cummings et al. 1995), taxon sampling (Graybeal 1998), and specific assumptions in the modelling of sequence evolution (Yang et al. 1994) can affect phylogenetic reconstruction. Biological processes, including the action of natural selection or genetic drift (Rieseberg et al. 1996; Maddison 1997; Satta et al. 2000; Martin and Burg 2002), hybridization (Naumov et al. 2000), and horizontal gene transfer (Bergthorsson et al. 2003) may cause the history of the genes/genomic regions under analysis to obscure the history of the taxa. However, to overcome the effect of analytical

and biological factors many researchers have attempted to address the problem by different ways of analysis (separate versus concatenated) of genome-wide data sets (Soltis et al. 1999; Baldauf et al. 2000; Moreira et al. 2000; Murphy et al. 2001). This may provide exceptional power not only in testing specific phylogenetic hypotheses but also in the precise reconstruction of the historical associations of all taxa analysed to recover the species tree.

Incongruent phylogenetic relationships among poplar species have challenged evolutionary biologists for more than a decade. A phylogenetic analysis of poplars using 76 morphological traits of buds, leaves, inflorescences, flowers and fruits supported the monophyly of all sections except for *Tacamahaca*, which resolved into two paraphyletic groups (Eckenwalder 1996). However, phylogenetic analysis based on RFLP data suggested that the section *Tacamahaca* is polyphyletic and mitochondrial and chloroplast restriction site analysis of four *Populus* species suggested a polyphyletic relationship for species in the section *Aigeiros* as well (Barrett et al. 1993; Rajora and Dancik 1995). Moreover, microsatellite based analysis (Rahman and Rajora 2002) revealed a close genetic relatedness between *P. deltoides* (section *Aigeiros*) and *P. trichocarpa* (section *Tacamahaca*) as well as between *P. balsamifera* (section *Tacamahaca*) and *P. nigra* (section *Aigeiros*) suggesting a polyphyly of the sections *Tacamahaca* and *Aigeiros*. Phylogenetic analyses of the genus *Populus* based on three non-coding regions of cpDNA *trnT-trnF* (intron of *trnL*, and intergenic regions of *trnT-trnL* and *trnL-trnF*) showed paraphyly for the section *Populus* (or monophyly with the exception of *P. nigra*) and *Aigeiros* and polyphyly in section *Tacamahaca*(Hamzeh and Dayanandan 2004). Based on the *ITS1* and *ITS2* regions of nuclear rDNA, however, monophyly for section *Populus*

and *Aigeiros* and paraphyly in sections *Tacamahaca* was shown (Hamzeh and Dayanandan 2004). Nuclear genomic data based upon inter simple sequence repeats (ISSR; Hamzeh et al. 2006) suggested the monophyletic origin of section *Populus* (*Leuce*), a polyphyletic relationship among species of section *Aigeiros* and a paraphyletic origin for section *Tacamahaca*.

Populus nigra could probably be considered as the most controversial species in the genus and its placement in any of the sections has been questioned. The cpDNA RFLP analysis of *P. nigra* showed similarity to species of the section *Populus*, but RFLP patterns of nuclear *rDNA* showed affinity to species of the section *Aigeiros* (Smith and Sytsma 1990). Interestingly, chloroplast DNA sequence data also showed a close affinity of *P. nigra* to species of the section *Populus*, although nuclear DNA sequence data suggested a close relationship between *P. nigra* and species of the section *Aigeiros* (Hamzeh and Dayanandan 2004). Allozyme and pollen competition studies suggested a closer evolutionary relationship between *P. deltoides* (section *Aigeiros*) and a member of section *Tacamahaca (P. maximowiczii)* than between *P. deltoides* and *P. nigra*, which are placed in the same section (Rajora 1989; Rajora and Zuffa 1990). Recently and in contrast to previous phylogenetic studies, ISSR data showed a close genetic relationship of *P. nigra* to species of the section *Tacamahaca*. This relationship is in agreement with various phenotypic traits, interfertility and the chemistry of bud exudates (Eckenwalder 1996) and serves as evidence for introgression between *P. nigra* and species of the section *Tacamahaca* (Hamzeh et al. 2006). Although the relatedness of species in the terminal clades of phylogenetic trees based on the nuclear and chloroplast nucleotide sequence data (Hamzeh and Dayanandan 2004) and nuclear genome ISSR markers

(Hamzeh et al. 2006) showed higher resolution than phylogenetic analysis based on RFLP data (Smith and Sytsma 1990), the evolutionary relationships among species within sections is still largely unresolved. Therefore, the observed low level of nucleotide sequence variability, abundant evidence for interspecific hybridization among the species, and incongruent molecular phylogenetic trees emphasises the need to use multiple of independent biparentally inherited loci to resolve the phylogenetic relationships among the species. In this way insight may be gained into the evolutionary history of these widely distributed forest tree and taxonomic units for this systematically controversial group of species may be defined.

In search for more variable and, therefore, more informative markers, we developed a novel group of molecular markers named SCISSRs or Sequence Characterised Inter Simple Sequence Repeats. Each SCISSR is a nucleotide sequence of a genomic region known as ISSR, which is flanked by two inverted microsatellites (SSR; Zietkiewicz et al. 1994). Wide coverage of the genome by microsatellites (SSRs are distributed throughout the genome) and the possibility of high variability of DNA sequences in regions near these repeats would make SCISSRs potentially valuable markers to detect evolutionary relationships between closely related species.

These regions of the genome can be amplified from single-primer PCR reactions where the primer is designed from di-or tri- nucleotide repeat motifs with a 3' anchoring sequence of one to three nucleotides (Gupta et al. 1994; Zietkiewicz et al. 1994). Since di- and tri-nucleotide SSRs are not found in the vascular plant chloroplast genome (Wolfe et al. 1998), the data generated through SCISSR analyses are representative of nuclear and therefore, both maternal and paternal lineages. This feature of SCISSR in conjunction

with their wide coverage of the genome and the high variability of the DNA sequences is valuable for revealing the comprehensive evolutionary history, such as introgression and hybrid lineages, of closely related species such as poplars.

The present study focusses on species of *Tacamahaca* and *Populus*, and the mainly North American *Aigeiros* sections in which evolutionary genetic relationships among species remain unresolved from previous studies (Hamzeh and Dayanandan 2004; Hamzeh et al. 2006). These include several ecologically and economically important species, some of which are widely used in plantations in North America and Europe. Tissue samples of *Populus* species with restricted geographical ranges were not available for the present study.

2. 2 MATERIALS AND METHODS

2. 2. 1 Taxon sampling and DNA extraction.

Taxon sampling was based on the classification of the genus *Populus* by Eckenwalder (1977a, b, 1996) and Dickmann and Stuart (1983). Fresh leaves were obtained from 21 taxa including 17 species representing three sections of the genus *Populus* (sections *Tacamahaca*, *Aigeiros*, and *Populus*), and two varieties of *P. deltoides* Bartr. ex Marshall. Depending on availability, we selected up to five individuals per species to discern the effect of intraspecific SCISSR variability. Two species of *Salix* (*S. integra* Thunb. and *S. nigra* Marshall) were used as outgroups. The list of species used in this study and the accession numbers are given in Table 2. 1. The total genomic DNA was extracted from frozen leaf tissue using the methods of Doyle and Doyle (1987) and Dayanandan et al. (1997).

2. 2. 2 Poplar SCISSR primer designing process.

The genomic DNA of selected individuals from different species was amplified in single-primer PCR reactions using five ISSR primers. The amplification reactions contained 230 μM dNTP, 2.5 mM $MgCl_2$, 0.4 μM of a given ISSR primer, 1 unit of Taq DNA polymerase, and 2.5μl buffer (0.2M Tris pH.9.5; 0.25 M KCl; 1mg/ml BSA, 5μl /ml tween 20) in a total volume of 25 μl (8 × 25 μl for each DNA/ primer combination). PCR amplification was performed in a Mastercycler gradient thermal cycler (Eppendorf, Westbury, New York, USA) with the touchdown thermal cycling protocol starting at 5°C above the optimum annealing temperature and a 1°C /cycle temperature decrease down to

the optimum annealing temperature. The remaining thermal cycling protocol was set to 94°C for 60 s, optimum annealing temperature for 30 s, and extension at 72°C for 60 s for a total of 38 cycles. The list of individuals, ISSR primers and their optimum annealing temperatures are given in Table 2. 2. PCR products were purified using the QIAquick PCR Purification Kit (QIAGEN Inc.- Mississauga, Ontario, Canada). Five µl of each eluted purified PCR product were electrophoresed on a 1% agarose gel with ethidium bromide (0.33µg/ml) at 3.5 v/cm for 90 minutes. ISSR bands were visualized and digitally documented using a GeneSnap 4.00–Gene Genius Bio Imaging System (Syngene, Frederick, Maryland, USA). The digital image files were analyzed using Gene Tools software (Syngene) and ISSR fragment sizes and quantities were estimated based on the MassRuler DNA Ladder Mix (Fermentas, Burlington, Ontario, Canada). Multiple ISSR bands of each purified PCR eluate were cloned randomly. ISSR bands were Ligated to the pGEM-T vector (Promega, Madison, Wisconsin, USA) using Promega 2 × Rapid Ligation Buffer for ligation of PCR products and Promega T4 DNA ligase (Promega, Madison, Wisconsin, USA). To calculate the appropriate amount of PCR product (insert) to include in the ligation reaction, the size and quantity of the biggest band of each purified PCR product on the agarose gel was considered in insert/vector ratio calculation. Five µl of each ligated DNA were used in the transformation of Subcloning Efficiency DH5α Competent Cells (Invitrogen Canada Inc.-Burlington, Ontario, Canada). Selection for transformants was performed on LB/ampicillin/IPTG/X-Gal plates. Six isolated white colonies per plate were picked and tested for positive transformation (versus false positives) by PCR amplification of recombinant plasmid DNA using pUC/M13 vector forward (5'-d(GTTTTCCCAGTCACGAC)-3') and reverse (5'-

d(CAGGAAACAGCTATGAC)-3') primer. Amplification reactions contained 230 μM dNTP, 2.5 mM $MgCl_2$, 0.2 μM of each primer, 1 unit of *Taq* DNA polymerase, and 2.5 μL buffer (0.2 M Tris PH.9.5; 0.25 M KCl; 1mg/ml BSA, 5 μl/ml Tween 20) in a total volume of 25 μL. PCR amplification was performed in a Mastercycler gradient thermal cycler (Eppendorf, Westbury, New York, USA) at 94°C for 60 s, 50°C for 30 s, and 72°C for 60 s for 35 cycles. 5 μl of each PCR product was electrophoresed on a 1% agarose gel with ethidium bromide (0.33μg/ml) at 7 v/cm for 45 minutes. Based on the gel electrophoresis result, positively transformed bacterial colonies containing various sizes of inserts from different plates were grown in 2.5 ml of LB/Amp liquid broth at 37°C for 16 h. Multiple recombinant plasmid DNAs were isolated from the bacterial cultures and purified using a QIAprep Spin Miniprep Kit (QIAGEN Inc.- Mississauga, Ontario, Canada) and sequenced at the McGill University and Genome Québec Innovation Centre Sequencing Platform (Montréal, Québec, Canada). Based on the sequences of the inserts, several sets of forward and reverse SCISSR primers were designed and synthesized (Operon Biotechnologies, Huntsville, Alabama, USA). The names and sequences of the SCISSR primers used in this study, the expected size of the SCISSR fragments (the size of the inserted ISSR fragments in Plasmid DNA), and the original genomic DNA/ ISSR combination from which they were cloned are given in Table 2. 3.

2. 2. 3 SCISSR data collection; PCR amplification and sequencing.

The genomic DNA of collected taxa was amplified using six sets of SCISSR primers (Table 2. 3). Amplification reactions contained 230 μM dNTP, 2.5 mM $MgCl_2$,

0.2 μM of each primer, 2 units of *Taq* DNA polymerase, and 2.5 μL buffer (0.2 M Tris PH.9.5; 0.25 M KCl; 1mg/ml BSA, 5 μl/ml Tween 20) in a total volume of 25 μL. PCR amplification was performed in a Mastercycler gradient thermal cycler (Eppendorf, Westbury, New York, USA) at 94°C for 60 s, 50°C for 30 s, and 72°C for 120 s for 45 cycles. PCR products were directly sequenced in both the forward and reverse directions for one of the SCISSR fragments (PIS6A1) and only in the forward direction for the rest of the fragments (this decision was made due to the good quality of the forward sequences, which would cover the full length of the fragments in most cases, and to avoid unnecessary sequencing of the large number of taxa used in this study) using the same corresponding PCR primer sets at the McGill University and Genome Québec Innovation Centre Sequencing Platform (Montréal, Québec, Canada). The chromatograms of the DNA sequencing results were processed and analyzed using the Staden software package (Staden 1979; http://staden.sourceforge.net). The assembled contigs of each SCISSR for all taxa were aligned using ClustalW (Thompson et al. 1994) multiple sequence alignment software. Aligned DNA sequences were imported into MacClade 4.0 software (Maddison and Maddison 2001) for verification and manual editing of the sequence alignments. Six nucleotide sequence data matrices were produced and analysed separately. Ultimately all of the nucleotide sequence data were combined and analysed together.

2. 2. 4 Separate phylogenetic analysis.

Using PAUP* v: 4.0 beta 8 (Swofford 2001), for each data set, 10^6 random trees were analyzed to obtain the frequency distribution of tree lengths in order to assess the

phylogenetic signal of the data matrix by calculating the skewness (g1) and kurtosis (g2). The nucleotide frequency distribution was investigated with a chi-square test of homogeneity of base distribution across sequences.

To avoid the loss of information that occurs when sequences are converted into distances, the discrete method of maximum parsimony, which operates directly on the nucleotide sequences, was used to reconstruct the tree (or trees) with the fewest evolutionary changes. Although the maximum likelihood estimate of phylogeny is also a discrete method, it is computationally time consuming and this has limited the application of the method to fairly small data sets (Page and Holmes 1998). Moreover, in practice, maximum parsimony and maximum likelihood trees can be very similar, especially when the expected amount of evolutionary change is small (Page and Holmes 1998). Therefore, the maximum parsimony optimality criterion was chosen to reconstruct the evolutionary relationships among *Populus* species.

Maximally parsimonious (MP) phylogenetic trees were reconstructed through heuristic search with equal character weights, gaps were treated as missing data, multistate taxa were interpreted as uncertainty, the starting tree was obtained via stepwise addition, and sequence addition was simple (random addition was not possible because of computing limitations). Tree-bisection reconnection (TBR) was used as the branch-swapping algorithm. The 50% majority rule consensus trees were obtained. The phylogenetic trees were rooted using *Salix* species as outgroups. Bootstrap analysis with a fast-heuristic search based on 1000 replicates was performed to assess the robustness of branches. Since data for all SCISSR fragments were not available for all taxa, the

taxonomic composition of each separate analyses is, by necessity, somewhat different from each other.

2. 2. 5 Combined phylogentic analysis.

To perform a total evidence analysis, all SCISSR data matrices were joined together producing a single concatenated matrix of six partitions.

Maximum parsimony analysis—Using PAUP* v: 4.0 beta 8 (Swofford 2001), 10^6 random trees were analyzed to obtain the frequency distribution of tree lengths in order to assess the phylogenetic signal of the combined data matrix by calculating the skewness (g1) and kurtosis (g2). The significance of incongruence among data partitions was measured by the incongruence length difference (ILD) test (Farris et al. 1994, 1995), which is implemented in PAUP* v: 4.0 beta 8 (Swofford 2001) as a partition-homogeneity test. However, in recognition of strengths and weaknesses of all three primary approaches to combine or not to combine data sets for phylogenetic estimation (total evidence, conditional combination, and separate analysis; Huelsenbeck et al. 1996), this study agree with Liu and Miyamoto (1999) that the homogeneity test should not be used to automatically justify the continued separation of data partitions. Therefore, to increase the "signal-to-noise ratio" in order to overcome the effect of analytical and biological factors in conflicting data (Murphy et al. 2001) phylogenetic analysis was performed under maximum parsimony criteria following the same steps used in each separate analysis mentioned above.

Bayesian analysis— The combined data matrix was also analyzed using Bayesian inference of phylogeny. Bayesian Inference (BI) was applied in order to overcome some

of the limitations such as Long-Branch Attraction (LBA) in the Maximum Parsimony (MP) method. BI differs from MP in being a model-based protocol, but possesses advantages over other model-based methods such as Maximum Likelihood (ML) in terms of its ability to use complex models of evolution with greater computational efficiency (Huelsenbeck et al. 2001, Nylander et al. 2004). A "GTR + I + G" evolutionary model of nucleotide substitution (General Time Reversible model with a proportion of invariable sites and a gamma-shaped distribution of rates across sites) was selected based on simultaneous evaluation of 56 models of nucleotide sequence evolution using Modeltest (Posada and Crandall 1998). Bayesian phylogenetic analysis were performed with the MrBayes version 3. 1 (Huelsenbeck and Ronquist 2001), using 1,000,000 generations with four chains (three chains ''heated'' using the default parameters) and sampling from the Markov Chain every 100^{th} generation. To determine when we have a good sample from the posterior probability distribution, using default option, two simultaneous, completely independent analyzes starting from different random tree were run. Trees from different runs were sampled and compared every 1000^{th} generations. To ensure sampling of topologies after chain convergence, the first 1000 trees were discarded as burn-in. Examination of the log-likelihood values and the observed consistency between runs suggested that this burn-in period was sufficiently long. A graphical representation of the Bayesian consensus tree was generated using PAUP* v: 4.0 beta 8 (Swofford 2001).

2. 3 RESULTS

2. 3. 1 Separate phylogenetic analyses.

Direct sequencing of purified PCR products of the each SCISSR fragments followed by ClustalW alignment of all sequences resulted in six data matrices with the length of 743, 410, 844, 1225, 907, and 639 characters (nucleotides and gaps) and 39, 43, 31, 41, 32, and 53 taxa with 68, 39, 48, 97, 128, and 51 parsimony informative characters for the SCISSR fragments PIS3A2, PIS5X1B4, PIS5A1, PIS6A1, PIS8A2, and PIS9A6 respectively (Appendices 2. 1, 2. 2, 2. 3, 2. 4, 2. 5, and 2. 6).

The frequency distribution of the length of 10^6 random trees yielded g1 = –0.509857, g2 = 0.198852 for PIS3A2, g1 = –0.351153, g2 = 0.051214 for PIS5X1B4, g1 = –1.050515, g2 = 1.484796 for PIS5A1, g1 = –0.559856, g2 = 0.326064 for PIS6A1, g1 = –0.643374, g2 = 0.435916 for PIS8A2, and g1 = –1.001011, g2 = 1.155198 for PIS9A6, suggesting a strong phylogenetic signal in these data matrices. The Chi-square test of homogeneity of base frequencies across taxa showed p = 1.0 in all cases, and the null hypothesis of homogeneous base distribution across sequences was accepted.

Individual SCISSR trees:

PIS3A2 tree— The maximum parsimony searches for SCISSR fragment PIS3A2 yielded 5250 equally parsimonious trees (Fig 2. 1: 50% majority rule consensus tree length 215; Consistency index, CI = 0.856; Retention index, RI = 0.890; Rescaled consistency index, RC = 0.761).

Based on MP analysis, all *Populus* species formed a strongly supported monophyletic group of two major sister clades (Fig. 2. 1). One clade comprised only species of section *Populus* and the second consisted of two distinct clades. one of which contained the species of section *Tacamahaca* plus one *P. nigra* individual (*P. nigra* 1) and the other included the members of section *Aigeiros* plus the remaining four individuals of *P. nigra* . Within the *Tacamahaca* lineage *P. maximowiczii*, *P. simonii*, and *P. laurifolia* (all Asiatic poplars) grouped together and made a sister clade to the clade containing the remaining members of the section. Finally *P. szechuanica* showed a close relationship to the North American *P. angustifolia* with a well supported internal node.

PIS5X1B4 tree— The maximum parsimony searches for SCISSR fragment PIS5X1B4 yielded 5879 equally parsimonious trees (Fig 2. 2: 50% majority rule consensus tree length 131; Consistency index, CI = 0.870; Retention index, RI = 0.928; Rescaled consistency index, RC = 0.808).

As with the PIS3A2 tree, all *Populus* species formed a strongly supported monophyletic group comprising two major clades (Fig. 2. 2). One clade comprised only species of section *Populus* and the second contained all the rest. With the exception of *P. maximowiczii* the rest of the species of section *Tacamahaca* clustered together. Within

this cluster there is a distinct group of all the North American species of section *Tacamahaca* (*P. balsamifera*, *P. trichocarpa*, and *P. angustifolia*) plus *P. szechuanica*, suggesting the close relatedness of this Asiatic species to the North American members of the section. However none of the relationships and internal nodes of this *Tacamahaca* clade was supported by bootstrap analysis. All the individuals of *P. nigra* however, grouped in a strongly supported clade with closer relationship to the members of *Tacamahaca* than to *P. deltoides* and *P. fremontii* in section *Aigeiros*.

PIS5A1 tree— The maximum parsimony searches for SCISSR fragment PIS5A1 yielded 234 equally parsimonious trees (Fig 2. 3 a and b: 50% majority rule consensus tree length 140; Consistency index, CI = 0.907; Retention index, RI = 0.917; Rescaled consistency index, RC = 0.832).

Due to a lack of sequence information for any of the *Salix* species used as the outgroup in this study, the maximum parsimony tree for this SCISSR is presented as an unrooted tree (Figs. 2. 3 a, and b). In this unrooted tree, a distinct lineage of section *Populus* was recognized although not all the individuals of the same species were grouped together. All the Asiatic species of section *Tacamahaca*, except for *P. szechuanica*, showed closer relationships to each other than to the North American members (*P. balsamifera* and *P. trichocarpa*) which also showed close relationships to each other. The phylogenetic relationships between *P. nigra* and the species of section *Tacamahaca* and *Aigeiros* remained unresolved since one of the *P. nigra* individuals (*P. nigra* 2) grouped with the lineage consisting of the Asiatic species of *Tacamahaca* while three other individuals (*P. nigra* 3, 4, and 5) had their own well supported lineage. These

two lineages were part of the five-descending branches polytomy, which also included the North American *Tacamahaca* species, the *P. angulata-P. fremontii* branch and the *P. nigra* 1 branch. Therefore, the phylogenetic position of *P. nigra* with regard to either section *Aigeiros* or *Tacamahaca* could not be determined.

PIS6A1 tree— The maximum parsimony searches for SCISSR fragment PIS6A1 yielded 415936 equally parsimonious trees (Fig 2. 4: 50% majority rule consensus tree length 208; Consistency index, CI = 0.899; Retention index, RI = 0.948; Rescaled consistency index, RC = 0.852).

The topology of the PIS6A1 tree is somewhat similar to the PIS3A2 tree. All *Populus* species formed a strongly supported monophyletic group consisting of two major clades (Fig. 2. 4). One clade included only species of section *Populus*, the second, however, consisted of three distinct clades. The first clade included most of the species of section *Tacamahaca*, although the phylogenetic relationships among the species remained unresolved. Within the second clade *P. maximowiczii*, *P. simonii*, and *P. laurifolia* (all Asiatic poplars) grouped together and made a strongly supported sister clade to the *P. nigra* clade (including all the studied individuals). Finally the third clade included all the members of section *Aigeiros* excluding *P. nigra* (*P. deltoides* and its varieties, *P. angulata* and *P. sargentii*, and *P. fremontii*).

PIS8A2 tree— The maximum parsimony searches for SCISSR fragment PIS8A2 yielded 1152 equally parsimonious trees (Fig 2. 5: 50% majority rule consensus tree

length 539; Consistency index, CI = 0.800; Retention index, RI = 0.691; Rescaled consistency index, RC = 0.553).

Similar to PIS3A2, PIS5X1B4, and PIS6A1 trees all *Populus* species formed a strongly supported monophyletic group comprising two major clades (Fig. 2. 5). One clade included only species of section *Populus* and the other contained the rest of species. Within this clade *P. nigra* showed a closer relationship to *P. simonii* (this relationship is strongly supported by bootstrap analysis), *P. maximowiczii*, *P. laurifolia* and *P. cathayana* of section *Tacamahaca* than to any member of section *Aigeiros*. Moreover, *P. szechuanica* showed a closer relationship to North American *P. trichocarpa* than to any Asiatic members of section *Tacamahaca*.

PIS9A6 tree— The maximum parsimony searches for SCISSR fragment PIS9A6 yielded 14030 equally parsimonious trees (Fig 2. 6: 50% majority rule consensus tree length 219; Consistency index, CI = 0.890; Retention index, RI = 0.876; Rescaled consistency index, RC = 0.780).

The analysis for PIS9A6 resulted in a tree with a very different topology to that of the other trees. Although all *Populus* species formed a strongly supported monophyletic group comprising again two also strongly supported major clades (Fig. 2. 6), one included only *P. nigra* while the other included all other species including those in section *Populus*. Within this clade species of the section *Populus* still form a closely related group with 100% majority rule consensus although the branch holding them together was not supported by bootstrap analysis. Moreover, species of section *Aigeiros* with the exception of *P. nigra* (*P. deltoides* and its varieties, *P. angulata* and *P. sargentii*,

and *P. fremontii*) formed a strongly supported clade. Similar to the PIS8A2 tree, *P. szechuanica* showed a closer relationship to North American *P. balsamifera* and *P. trichocarpa* of the section *Tacamahaca* rather than to other Asiatic members of the section.

2. 3. 2 Combined phylogenetic analysis.

Concatenation of all the six separate SCISSR matrices resulted in one combined data matrix with the length of 4768 characters (nucleotides and gaps) and 44 taxa with 427 parsimony informative characters. The frequency distribution of the length of 10^6 random trees yielded g1 = –0.539415, g2 = 0.318125 suggesting the presence of a phylogenetic signal in the data matrix. The partition homogeneity test showed significant heterogeneity levels between data partition with p = 0.01.

Maximum parsimony combined data tree— Maximum parsimony searches of six combined SCISSR matrices yielded 660 equally parsimonious trees (Fig. 2. 7: 50% majority rule consensus tree length 1553; Consistency index, CI = 0.800; Retention index, RI = 0.806; Rescaled consistency index, RC = 0.645).

Based on this analysis, all *Populus* species formed a strongly supported monophyletic group comprising two major well supported clades itself (with bootstrap analysis support of 85% and 75%; Fig. 2. 7). Multiple individuals of the same species clustered together in this tree suggesting that the level of intraspecific variation between individuals (presented by the branch length in the cluster of each species multiple individuals) did not overwhelm the interspecific phylogenetic relationships among the

species. One clade included all the species of section *Populus* indicating the monophyletic origin of this section, which will be referred to as the *Populus* lineage hereafter. The other, clade which will be referred to as the second clade after this, included all species from the two remaining sections, section *Tacamahaca* and section *Aigeiros*.

In the *Populus* linage *P. alba*, the only poplar member of the section *Populus*, appeared in a basal position to the other species of the section (all aspens) with strong bootstrap support (99%). *Populus tremuloides* (North American trembling aspen) showed closer relationship to *P. grandidentata* (another North American aspen) than to *P. tremula* (European aspen), which showed closer affinity to *P. davidiana* (Korean aspen). Although all these relationships maintained a high percentage of occurrence in the 50% majority rule consensus MP tree, they were not supported by bootstrap analysis.

In the second clade two major clusters can be recognized. One strongly supported (with 83% bootstrap support) group consisted of all the North American cottonwoods of section *Aigeiros* while the other group included all the species of section *Tacamahaca* and the controversial member of section *Aigeiros*, *P. nigra*. Within this group, *P. laurifolia* occupied a basal position to a cluster consisting of two sister clades, *P. nigra* and *P. simonii* - *P. maximowizcii*. The rest of the species of section *Tacamahaca* (*P. trichocarpa*, *P. balsamifera*, *P. angustifolia*, *P. tristis*, *P. cathayana*, and *P. szechuanica*) grouped together with a fairly good bootstrap support of 60%. However, the relationships among these species remained unresolved.

Bayesian phylogenetic inference of combined data—The results of the Modeltest analysis showed that GTR+ I+ G nucleotide substitution model (General Time Reversible model with a proportion of invariable sites and a gamma-shaped distribution of rates across sites) was the most suitable model for the combined data. The parameters of the model were: base frequencies: A = 0.2789, C = 0.2210, G = 0.1984, T = 0.3017; rate matrix: (A-C) = 0.6682, (A-G) = 1.8935, (A-T) = 0.6654, (C-G) = 0.7603, (C-T) = 1.3396, (G-T) = 1.0; among-site rate variation: proportion of invariable sites (I) = 0.3743; variable sites (G): gamma distribution shape parameter= 0.8488.

The 50% majority rule consensus of 9001 trees derived from Bayesian analysis of combined data with the described model parameters retained a tree (Fig. 2. 8) with a topology congruent with the 50% majority rule consensus tree obtained from parsimony analysis with few minor differences in intraspecific relationship among multiple individual. As with the MP analysis, the Bayesian analysis also robustly confirmed the monophyletic origin of the genus *Populus* comprising two major well supported clades itself with 100% posterior probability for each clade. One clade included all the species of section *Populus* indicating the monophyletic origin of this section, and the other clade included all species from the two remaining sections, section *Tacamahaca* and section *Aigeiros*.

The closer relationship between *Populus tremuloides* (North American trembling aspen) and *P. grandidentata* (another North American aspen) than to *P. tremula* (European aspen), which was suggested in MP tree, was supported with high (74%) posterior probability. Moreover, in the second clade the same two major clusters as the MP tree could be recognized too. One group consisted of all the North American

cottonwoods of section *Aigeiros* while the other group included all the species of section *Tacamahaca* and *P. nigra*. Within this group, *P. laurifolia* occupied a basal position to a cluster consisting of two sister clades, *P. nigra* and *P. simonii* - *P. maximowizcii*. Finally, the rest of the species of section *Tacamahaca* (*P. trichocarpa*, *P. balsamifera*, *P. angustifolia*, *P. tristis*, *P. cathayana*, and *P. szechuanica*) grouped together. However, contrary to low bootstrap support of these clades in MP tree, the credibility of all of these relationships was strongly supported with 100% posterior probability in Bayesian analysis.

2. 4 DISCUSSION

The question of whether to combine or not to combine data sets for phylogenetic analysis has remained controversial. There are three main approaches to the problem of combining data in phylogenetic analysis. First the 'always combine' or 'total evidence' approach of Kluge (1989, 1998), which propose that phylogenetic analysis should be performed on a combined data set using all possible evidence because it maximizes the "informativeness' and 'explanatory power' of the character data used in the analysis. The total evidence approach employs the null hypothesis that there are no significant differences or partitions within the data sets. This is based on the philosophical justification that there is one evolutionary history for the taxa under investigation which should be reflected in all data partitions. Second the 'separate analysis' approach (Miyamoto and Fitch 1995), which estimates trees from each data partition individually and examines the congruence of these trees. Third the 'conditional combination' approach (Bull et al. 1993; de Queiroz 1993; Rodrigo et al. 1993; de Queiroz et al. 1996), which performs a statistical test of the null hypothesis of data homogeneity. If this null hypothesis is rejected, one interpretation could be that different trees underlie each data partition; a situation that can be encountered with ancestral polymorphism, hybridization, or horizontal gene transfer. In this case combining the data in a single analysis is not warranted. However, Cunningham (1997) performed various statistical tests to explore the relationship between incongruence and phylogenetic accuracy. He found that with heterogeneity value of $p = 0.01$, combining data might still improve phylogenetic accuracy. As noted by Huelsenbeck et al. (1996) estimates from each data partition are

based on a smaller sample of characters than the combined analysis and will be subject to more sampling error than combined analysis. Since the overall accuracy of the combined data may be increased by a large number of characters applied to parts of the tree unaffected by the mismatch (Wiens 1998), combining data sets despite high levels of character incongruency may still be advantageous. Therefore, we adopted the position that data sets should be analysed both separately to assess the distribution, nature and extent of conflict among data sets, and combined simultaneously to potentially increase the descriptive efficiency and explanatory power of the data (Eernisse and Kluge 1993).

The null hypothesis of homogeneity among our data partition was rejected and MP trees recovered from single SCISSR data set in separate analysis were incongruent and they exhibited topological differences. However, the degree of conflict among trees was relatively minor. The maximum parsimony and Bayesian inference based phylogenetic trees of combined data resulted in similar and congruent phylogenetic topologies. Therefore, in order to discuss the evolutionary patterns in *Populus*, tree topology similar to the 50% majority rule MP or Bayesian tree derived from combined six SCISSR data sets, which will be referred to as the combined tree after this was considered to be the most plausible hypotheses for the phylogenetic relationships within the genus *Populus* (Fig 2. 7 and 2. 8). The robustness and credibility of the clades and relationships among species was evaluated by bootstarp value and posterior probability of the clades in MP and Bayesian trees respectively.

The combined tree (Fig. 2. 7, and 2. 8) showed monophyly of species of genus *Populus*, polyphyletic relationship among species of section *Aigeiros* and paraphyly of section *Tacamahaca*. The strong support (85% bootstrap value and 100% posterior

probability support) for the monophyly of the genus *Populus* is in accordance with previous study based on nuclear genomic data, which supported the monophyly of the genus *Populus* (Hamzeh et al. 2006). Moreover, the relationships among species within the section agree with the ISSR derived dendrogram (Hamzeh et al. 2006). *Populus alba* (white poplar), the only 'poplar' member of the section in this study, occupied a strong basal and distinct position compared to the rest of the species, reflecting the phenotypic uniqueness and morphological differences of this species. The other clade of the section was separated into two sister groups of Eurasian (*P. davidiana* and *P. tremula*) and North American (*P. grandidentata* and *P. tremuloides*) aspens.

Although the paraphyly of section *Tacamahaca* (Hamzeh and Dayanandan 2004; Hamzeh et al. 2006) and polyphyly of section *Aigeiros* (Hamzeh et al. 2006) have also been suggested before, in the combined SCISSR tree it is only the placement of *P. nigra* in the clade comprising all the species of section *Tacamahaca* that makes section *Aigeiros* polyphyletic and section *Tacamahaca* paraphyletic lineages. Otherwise, section *Aigeiros* with strong 83% bootstrap support and 100% posterior probability support and section *Tacamahaca* with 100% occurrence in all 660 equally parsimonious trees (though with <50% bootstrap support) and 100% posterior probability support would be perfect monophyletic sections.

The placement of *P. nigra* (the most controversial species in the genus) in any of the sections remained disputable. The analyses of the chloroplast genome related it to section *Populus* (Smith and Sytsma 1990; Hamzeh and Dayanandan 2004). Some nuclear genomic data, however, as well as morphological studies, have associated it with section *Aigeiros* (Smith and Sytsma 1990; Hamzeh and Dayanandan 2004) and others

have suggested its affinity with section *Tacamahaca* (Hamzeh et al. 2006). The evolutionary affinity between *P. nigra* and species of section *Tacamahaca* has been supported by interfertility (*P. nigra* x *P. trichocarpa* and *P. nigra* x *P. laurifolia*; Dickmann and Stuart 1983) and bud exudate data (Eckenwalder 1996). Based on the MP and Bayesian tree derived from six combined SCISSR data, *P. nigra* showed a close relationship with three Asiatic member of section *Tacamahaca*: *P. maximowiczii*, *P. simonii*, and *P. laurifolia*. Although this relationship is not supported by bootstrap analysis, the 100% occurrence of internal nodes and connecting branches in the 50% majority rule consensus MP tree with 100% posterior probability support in Bayesian tree, indicating the existence of valid genetic relationships among these species. However, the evidence from classical morphological, chemical, crossing (*P. nigra* can hybridize with *P. deltoides* and *P. fremontii* from section *Aigeiros* as well), and pathogenic studies do not consistently favour the placement of black poplar in either section *Aigeiros*, *Tacamahaca* or section *Populus*. Moreover, the close relationship between *P. maximowiczii*, *P. simonii*, and *P. laurifolia* compared to the rest of the *Tacamahaca* species has also been suggested previously (Hamzeh and Dayanandan 2004).

Contrary to an ISSR based distance tree (Hamzeh et al. 2006) and cpDNA nucleotide sequence data (Hamzeh and Dayanandan 2004), which linked *P. tristis* to North American cottonwoods of section *Aigeiros*, in the combined SCISSR tree *P. tristis* clustered with a group of North American as well as Asiatic species of section *Tacamahaca* (*P. balsamifera*, *P. trichocarpa*, *P. angustifolia*, *P. szechuannica*, and *P. cathayana*). The relationships among this group of species was not resolved, although

their cluster had bootstrap support of 60% (reasonably higher that the 50% threshold) and 100% posterior probability support.

Phylogenetic relationships among North American cottonwoods of section *Aigeiros* based on SCISSR data showed that *P. sargentii*, commonly known as plains cottonwood, which is often classified as *P. deltoides* var. *occidentalis* Rydb. or *P. deltoides* ssp *monilifera* (Ait.) Ecken. is closely related to *P. deltoides*. However, *P. angulata*, which is also classified as *P. deltoides* var. *angulata* Ait. (Dickmann and Stuart 1983; Eckenwalder 1996) showed a close relationship to *P. fremontii* rather than to *P. deltoides*. All the internal nodes of these relationships are well supported by bootstrap analysis and 98-100% posterior probability.

The evolutionary genetic relationships between sections *Aigeiros* and *Tacamahaca* have long been suspected (Rajora 1989; Rajora and Zuffa 1990; Hamzeh and Dayanandan 2004; Hamzeh et al. 2006) and species of these two sections are known to be freely inter-fertile (Zsuffa 1975; Eckenwalder 1984a; Ronald 1982; Rajora 1989). Since the degree of relative inter-fertility is assumed to be a reflection of genetic relatedness (Stettler et al. 1996 a; Dickmann et al. 2001), it can form a basis to infer taxonomic affinities. This is consistent with the result derived from combined SCISSR data, which put a clade consisting of species of section *Tacamahaca* plus *P. nigra* in a sister position to the *Aigeiros* clade (all the species of the section except *P. nigra* or black poplar). Moreover, the basal node of this close relationship is supported by a strong bootstrap value of 75% and posterior probibility of 100%.

In conclusion we have used concatenations of six nuclear genome based SCISSR data sets to assess genetic relationships among species of the genus *Populus*, a closely

related group of species, in which other genetic markers showed limited success in resolving the evolutionary relatedness. The phylogenetic tree based on six combined SCISSR markers showed higher resolution than RFLP, ISSR, single SCISSR, and DNA sequence based phylogenetic trees. However, most of the internal nodes depicting speciation events (and therefore, phylogenetic relationships among species) either were not resolved or supported by bootstrap value and posterior probability.

The results of our current study as well as previous ones (Hamzeh and Dayanandan 2004; Hamzeh et al. 2006) is a good illustration of the fact that analyses based on single or small number of genes or genomic data provide insufficient evidence for establishing phylogenetic hypotheses (Rokas et al. 2003 b). Since there may be no good indicator of the phylogenetic informativeness of genes, regardless of the source of incongruence, it is only through analyses of a large amount of sequence data that a confident phylogenetic reconstruction can be obtained (Rokas et al. 2003 b). However, the amount of sequence information needed to resolve specific relationships will depend on the particular evolutionary history under study. For example, in closely related species such as poplars, branches depicting speciation events separated by short intervals would be much harder to resolve and may need more data compared to branches depicting speciation events separated by long time intervals in distantly related taxa. Therefore, in order to gain insight into the evolutionary history of species of the genus *Populus*, we need larger concatenated genome-wide sampling of independently evolving and unlinked data sets for precise reconstruction of phylogenetic association of all the species.

Table 2. 1 Taxa of genus *Populus* L. and outgroups sampled for this study, their common names, voucher numbers, source, and geographic origin (the classification system of *Populus* follows Eckenwalder 1977a, b, 1996, and Dickmann and Stuart, 1983; AP, Alberta Pacific Forest Products Collection; MRNFQ, Le Ministere des Ressources Naturelles et de la Faune du Quebec; MBG, Montreal Botanical Garden; MAM, Morden Research Station and the Arboretum, Manitoba; MAQ, Morgan Arboretum, Quebec).

Taxon	Common name	Voucher number	Source	Geographic origin
Sect. *Populus (Leuce* Duby) subsect. Albidae				
P. alba L.	White poplar	MH-002, MH-003, MH-004	AP	Eurasia & N. Africa
Sect. *Populus* (*Leuce* Duby) subsect. Trepidae				
P. davidiana Schneid.	Korean aspen	MH-007, MH-008, MH-010, MH-011, MH-012	AP	Asia
P. grandidentata Michx.	Bigtooth aspen	MH-013, MH-014, MH-015, MH-016	MRNFQ	North America
P. tremuloides Michx.	Trembling aspen	MH-025, MH-026, MH-027	AP	North America
P. tremula L.	European aspen	MH-019, MH-021	AP	Eurasia & N. Africa
Sect. *Tacamahaca* Spach.				
P. angustifolia James	Narrowleaf cottonwood	MH-059	MAM	North America
P. balsamifera L.	Balsam poplar	MH-054, MH-087 MH-092	AP	North America
P. cathayana Rehder		MH-060	MA	Asia
P. laurifolia Ledeb.	Laurel poplar	MH-061, MH-062	MA	Asia
P. maximowiczii A. Henry	Japanese poplar	MH-031, MH-032	MRNFQ	Asia
P. simonii Carriere	Simon poplar	MH-065	MBG	Asia
P. szechuanica C. K. Schneid.	Szechuan poplar	MH-066	MBG	Asia
P. trichocarpa Torr. & Gray ex Hook	Black cottonwood	MH-040, MH-041	MRNFQ	North America
P. tristis Fisch.	Himalayan balsam poplar	MH-067	MBG	Asia
Sect. *Aigeiros* Duby				
P. deltoides Bartr. ex Marsh.	Eastern cottonwood	MH-048, MH-050, MH-052	AP	North America
P. deltoides var . *angulata* Ait.	Alamo cottonwood	MH-071	MAM	North America
P. sargentii Dode (*P. deltoides* var. *occidentalis* Rydb.)	Plains cottonwood	MH-070	MAM	North America
P. fremontii S. Watson	Fremont cottonwood	MH-063	MAM	North America
P. nigra L.	Black poplar	MH-043, MH-044, MH-045, MH-046, MH-047	MRNFQ	Eurasia
Outgroup taxa				
Salix nigra Marshall		MH-S-001	MAQ	
Salix integra		MH-S-002	MAQ	

Table 2. 2 Sequences of ISSR primers, optimum annealing temperature for each ISSR primer, and the voucher number of selected poplar species individuals. * Primers from Wolfe et al. (1998) and ** primers from Hamzeh et al. (2006).

Primer sequence	Optimum annealing temperature (°C)	Species Voucher number
**(ATC) 10-RC	55	MH-048, MH-067
*(CA) 6-RG	45	MH-007, MH-060, MH-066
*(CT) 8-RG	45	MH-021, MH-054, MH-066
*(CTC) 4-RC	45	MH-003, MH-014, MH-049
**(AGC) 10-RC	60	MH-007, MH-070

Table 2. 3 Names and sequences of the SCISSR primers used in this study, the expected size of the SCISSR fragments (the size of the inserted ISSR fragments in Plasmid DNA), and the original genomic DNA/ ISSR PCR combination from which they were cloned.

SCISSR Fragment and Primer Name	Forward and Reverse Primer Sequence	Origin of Cloned Band (Species Voucher # / ISSR Combination)	Expected Amplified Fragment Size
PIS3A2	F: 5'-TCATCGCTCCAAGGTACAAAC-3' R:5'-ACTAAGCAGCAGCCCAATTC-3'	MH-067/ (ATC) 10-RC	~740 bp
PIS5X1B4	F:5'-GGCAACACCTGCTGTTTTAAG-3' R:5'-ACACAAGATTGGAAGCTCCTG-3'	MH-007/ (CA) 6-RG	~460 bp
PIS5A1	F:5'-GCTATTAACATATGTCGACGGAG-3' R:5'-CATGGACATAAACAACCATGAG-3'	MH-066/ (CA) 6-RG	~1800 bp
PIS6A1	F:5'-TGCATCCACAGAATGGAAATC-3' R:5'-ATGTGGCAATGAACAAAGTCG-3'	MH-021/ (CT) 8-RG	~1250 bp
PIS8A2	F:5'-TTTTATCCCTGCCACCTTTAC-3' R:5'-CTCATAGCCCGTCTCATCATC-3'	MH-014/ (CTC) 4-RC	~1000 bp
PIS9A6	F:5'-AAGGCACGAAGAGTTCCGTAG-3' R:5'-GCGAAAGGAAAGCCCTGA-3'	MH-007/ (AGC) 10-RC	~650 bp

Figure 2. 1 The 50% majority rule consensus tree of 5250 equally parsimonious trees (tree length 215; CI = 0.856) based on SCISSR fragment PIS3A2 from *Populus* species. Numbers above branches show the frequency of occurrence in 50% majority rule consensus tree and numbers below branches are bootstrap values >50% based on 1000 replicates. Branches without numbers had bootstrap values <50%. Numbers in brackets show branch lengths (number of nucleotide substitution).

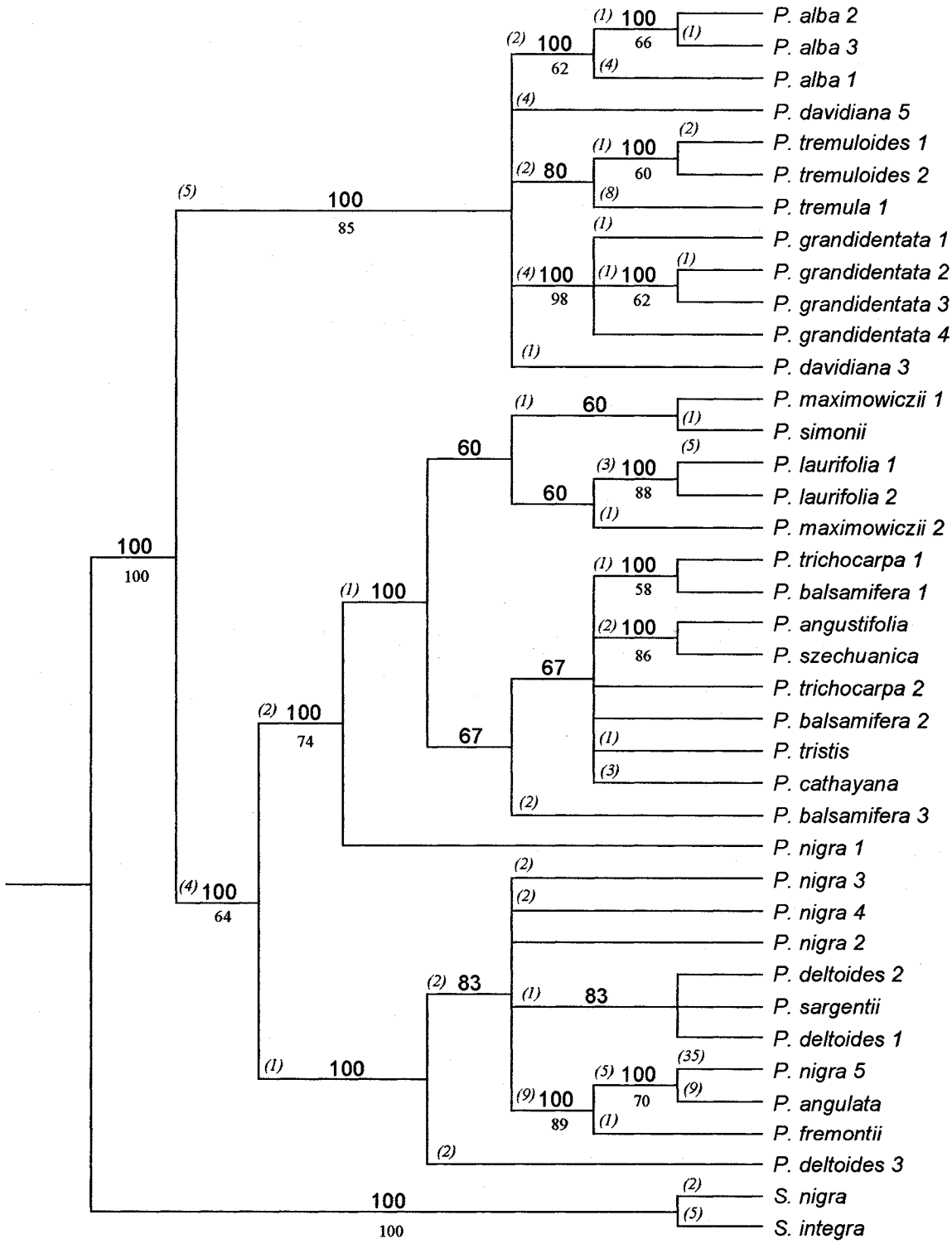

P. alba 2
P. alba 3
P. alba 1
P. davidiana 5
P. tremuloides 1
P. tremuloides 2
P. tremula 1
P. grandidentata 1
P. grandidentata 2
P. grandidentata 3
P. grandidentata 4
P. davidiana 3
P. maximowiczii 1
P. simonii
P. laurifolia 1
P. laurifolia 2
P. maximowiczii 2
P. trichocarpa 1
P. balsamifera 1
P. angustifolia
P. szechuanica
P. trichocarpa 2
P. balsamifera 2
P. tristis
P. cathayana
P. balsamifera 3
P. nigra 1
P. nigra 3
P. nigra 4
P. nigra 2
P. deltoides 2
P. sargentii
P. deltoides 1
P. nigra 5
P. angulata
P. fremontii
P. deltoides 3
S. nigra
S. integra

Figure 2. 2 The 50% majority rule consensus tree of 5879 equally parsimonious trees (tree length 131; CI = 0.870) based on SCISSR fragment PIS5X1B4 from *Populus* species. Numbers above branches show the frequency of occurrence in 50% majority rule consensus tree and numbers below branches are bootstrap values >50% based on 1000 replicates. Branches without numbers had bootstrap values <50%. Numbers in brackets show branch lengths (number of nucleotide substitution).

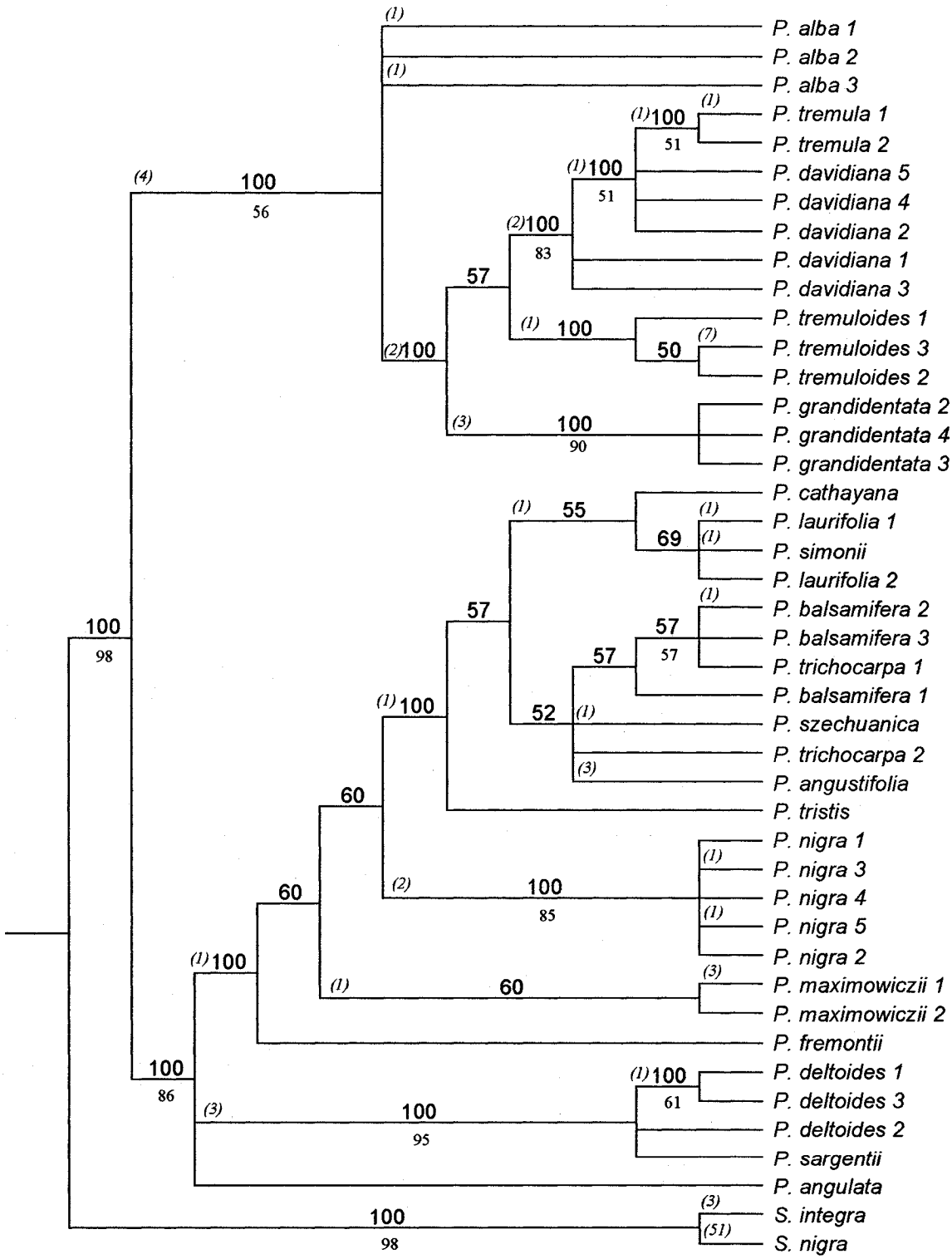

P. alba 1
P. alba 2
P. alba 3
P. tremula 1
P. tremula 2
P. davidiana 5
P. davidiana 4
P. davidiana 2
P. davidiana 1
P. davidiana 3
P. tremuloides 1
P. tremuloides 3
P. tremuloides 2
P. grandidentata 2
P. grandidentata 4
P. grandidentata 3
P. cathayana
P. laurifolia 1
P. simonii
P. laurifolia 2
P. balsamifera 2
P. balsamifera 3
P. trichocarpa 1
P. balsamifera 1
P. szechuanica
P. trichocarpa 2
P. angustifolia
P. tristis
P. nigra 1
P. nigra 3
P. nigra 4
P. nigra 5
P. nigra 2
P. maximowiczii 1
P. maximowiczii 2
P. fremontii
P. deltoides 1
P. deltoides 3
P. deltoides 2
P. sargentii
P. angulata
S. integra
S. nigra

Figure 2. 3 a The 50% majority rule consensus tree of 234 equally parsimonious trees (tree length 140; CI = 0.907) based on SCISSR fragment PIS5A1 from *Populus* species. Numbers above branches show the frequency of occurrence in 50% majority rule consensus tree.

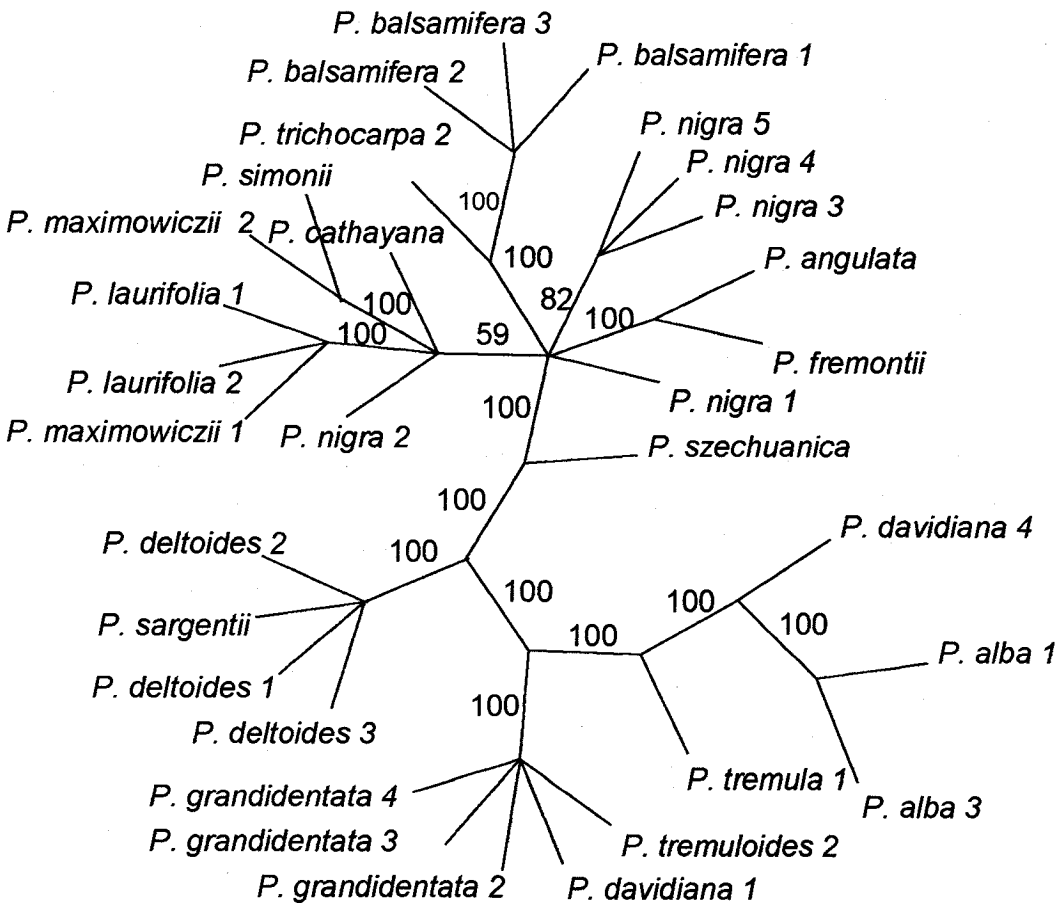

P. balsamifera 3
P. balsamifera 1
P. balsamifera 2
P. trichocarpa 2
P. nigra 5
P. nigra 4
P. simonii
P. nigra 3
P. maximowiczii 2
P. cathayana
P. angulata
P. laurifolia 1
P. fremontii
P. laurifolia 2
P. nigra 1
P. maximowiczii 1
P. nigra 2
P. szechuanica
P. davidiana 4
P. deltoides 2
P. sargentii
P. alba 1
P. deltoides 1
P. deltoides 3
P. tremula 1
P. alba 3
P. grandidentata 4
P. grandidentata 3
P. tremuloides 2
P. grandidentata 2
P. davidiana 1
100
100
100
82
100
100
59
100
100
100
100
100
100
100
100

Figure 2. 3 b The 50% majority rule consensus tree of 234 equally parsimonious trees (tree length 140; CI = 0.907) based on SCISSR fragment PIS5A1 from *Populus* species. Numbers below branches are bootstrap values >50% based on 1000 replicates. Branches without numbers had bootstrap values <50%. Numbers in brackets show branch lengths (number of nucleotide substitution).

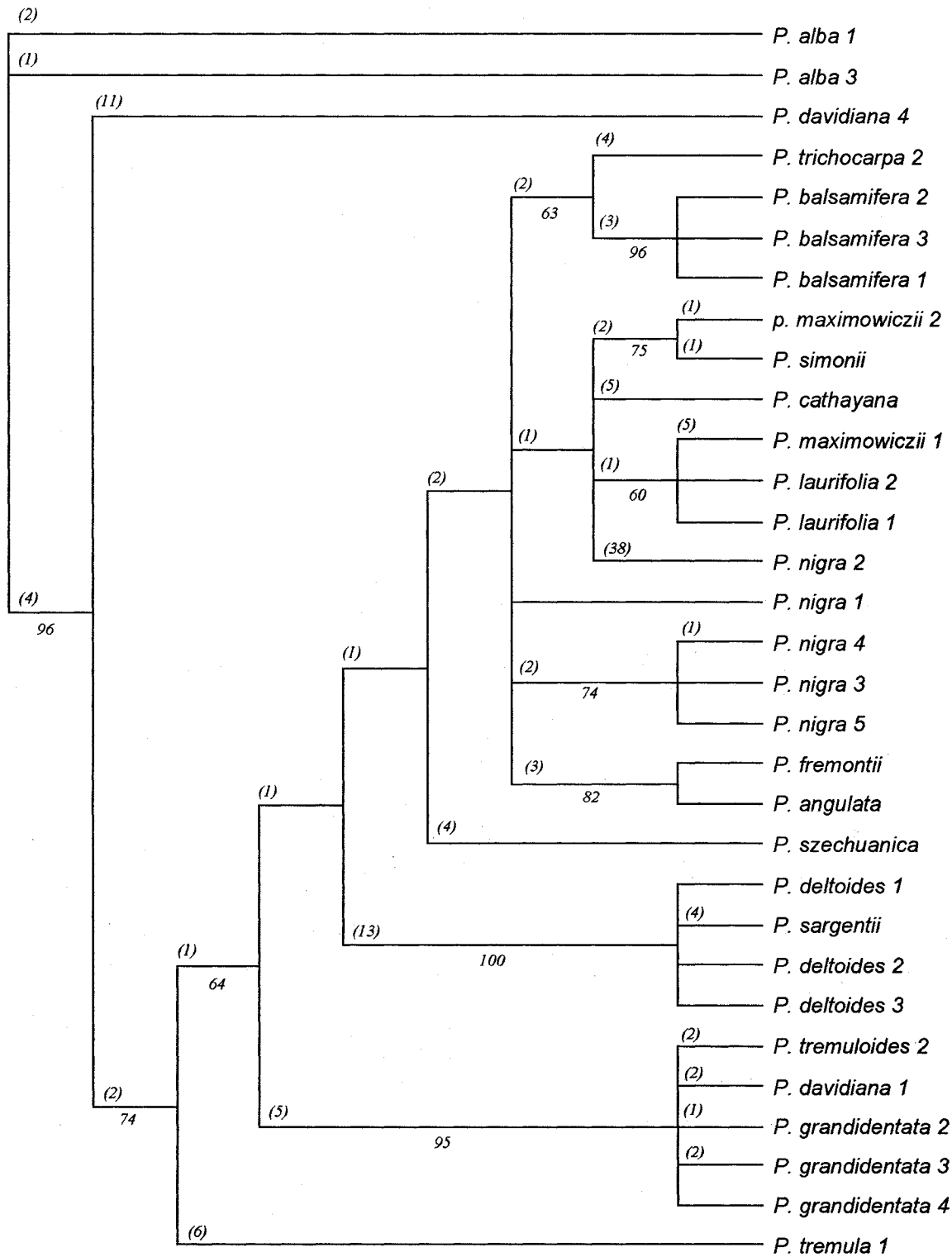

(2)
(1)
(11)
(4)
96
(2)
74
(1)
64
(6)
(1)
(5)
95
(1)
(13)
100
(2)
(4)
(2)
63
(3)
96
(1)
(2)
(1)
60
(2)
74
(3)
82
(4)
(2)
75
(1)
(5)
(1)
(38)
(1)
(5)
(2)
(2)
(1)
P. alba 1
P. alba 3
P. davidiana 4
P. trichocarpa 2
P. balsamifera 2
P. balsamifera 3
P. balsamifera 1
p. maximowiczii 2
P. simonii
P. cathayana
P. maximowiczii 1
P. laurifolia 2
P. laurifolia 1
P. nigra 2
P. nigra 1
P. nigra 4
P. nigra 3
P. nigra 5
P. fremontii
P. angulata
P. szechuanica
P. deltoides 1
P. sargentii
P. deltoides 2
P. deltoides 3
P. tremuloides 2
P. davidiana 1
P. grandidentata 2
P. grandidentata 3
P. grandidentata 4
P. tremula 1

Figure 2. 4 The 50% majority rule consensus tree of 415936 equally parsimonious trees (tree length 208; CI = 0.899) based on SCISSR fragment PIS6A1 from *Populus* species. Numbers above branches show the frequency of occurrence in 50% majority rule consensus tree and numbers below branches are bootstrap values >50% based on 1000 replicates. Branches without numbers had bootstrap values <50%. Numbers in brackets show branch lengths (number of nucleotide substitution).

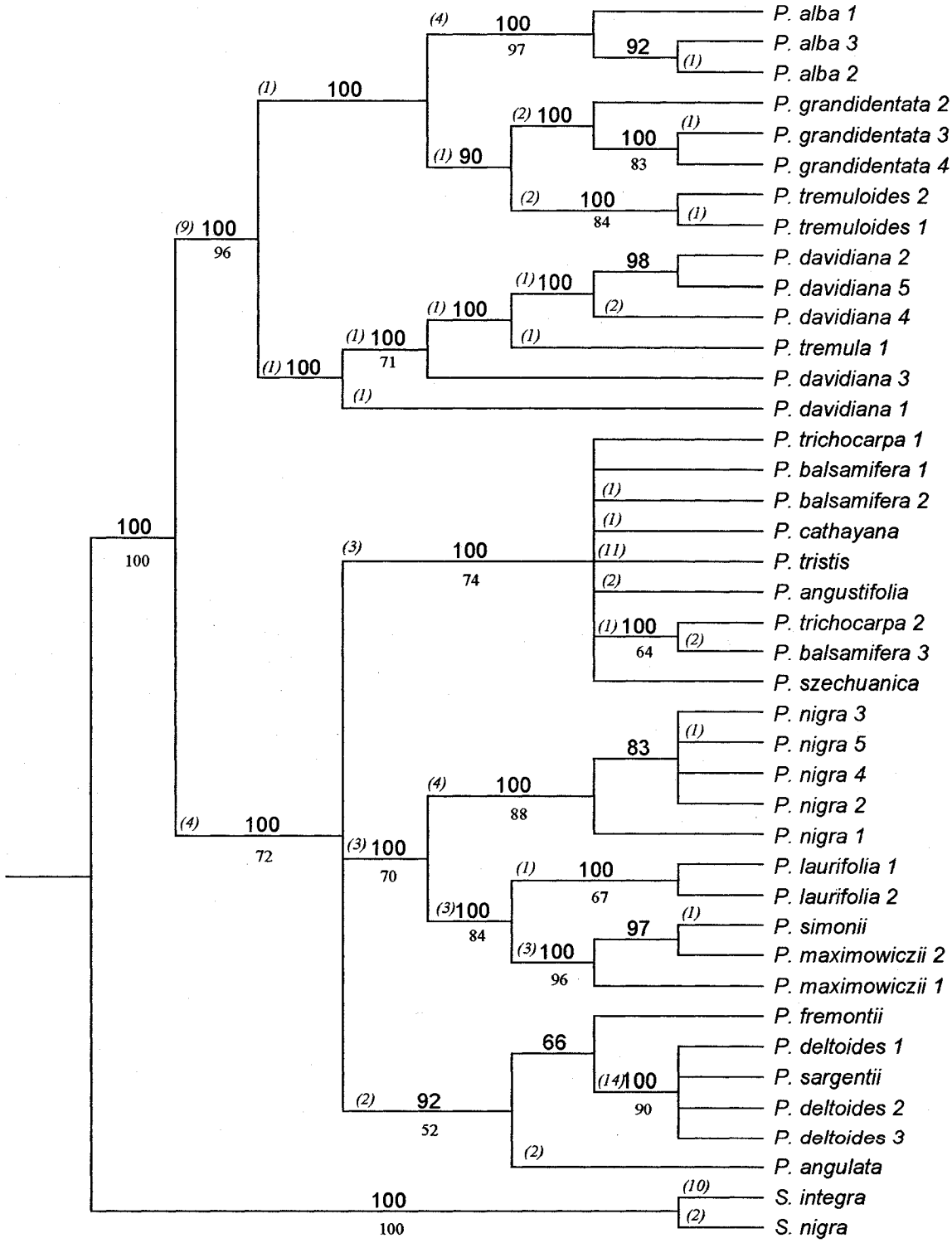

P. alba 1
P. alba 3
P. alba 2
P. grandidentata 2
P. grandidentata 3
P. grandidentata 4
P. tremuloides 2
P. tremuloides 1
P. davidiana 2
P. davidiana 5
P. davidiana 4
P. tremula 1
P. davidiana 3
P. davidiana 1
P. trichocarpa 1
P. balsamifera 1
P. balsamifera 2
P. cathayana
P. tristis
P. angustifolia
P. trichocarpa 2
P. balsamifera 3
P. szechuanica
P. nigra 3
P. nigra 5
P. nigra 4
P. nigra 2
P. nigra 1
P. laurifolia 1
P. laurifolia 2
P. simonii
P. maximowiczii 2
P. maximowiczii 1
P. fremontii
P. deltoides 1
P. sargentii
P. deltoides 2
P. deltoides 3
P. angulata
S. integra
S. nigra

Figure 2. 5 The 50% majority rule consensus tree of 1152 equally parsimonious trees (tree length 539; CI = 0.800) based on SCISSR fragment PIS8A2 from *Populus* species. Numbers above branches show the frequency of occurrence in 50% majority rule consensus tree and numbers below branches are bootstrap values >50% based on 1000 replicates. Branches without numbers had bootstrap values <50%. Numbers in brackets show branch lengths (number of nucleotide substitution).

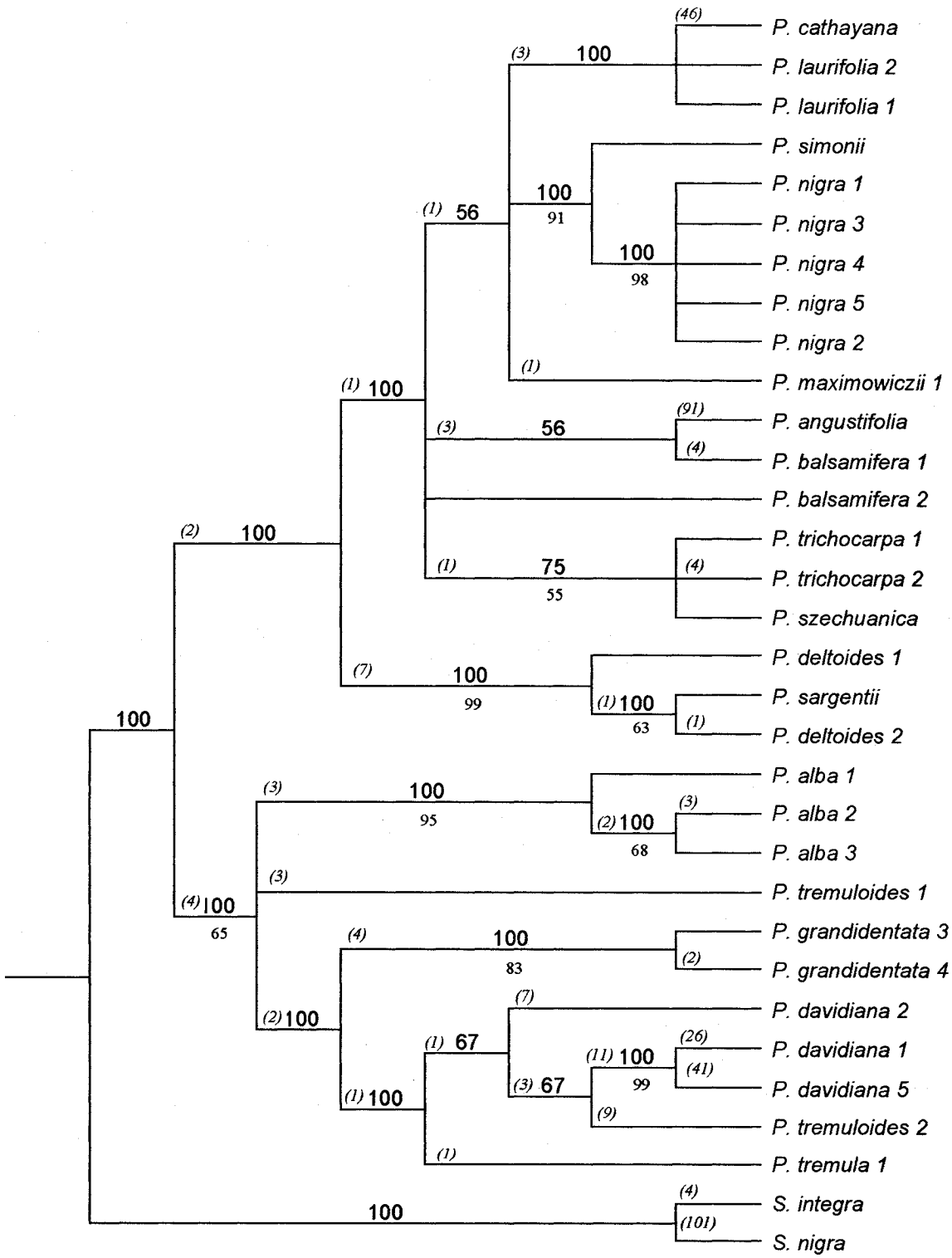

P. cathayana
P. laurifolia 2
P. laurifolia 1
P. simonii
P. nigra 1
P. nigra 3
P. nigra 4
P. nigra 5
P. nigra 2
P. maximowiczii 1
P. angustifolia
P. balsamifera 1
P. balsamifera 2
P. trichocarpa 1
P. trichocarpa 2
P. szechuanica
P. deltoides 1
P. sargentii
P. deltoides 2
P. alba 1
P. alba 2
P. alba 3
P. tremuloides 1
P. grandidentata 3
P. grandidentata 4
P. davidiana 2
P. davidiana 1
P. davidiana 5
P. tremuloides 2
P. tremula 1
S. integra
S. nigra

Figure 2. 6 The 50% majority rule consensus tree of 14030 equally parsimonious trees (tree length 219; CI = 0.890) based on SCISSR fragment PIS9A6 from *Populus* species. Numbers above branches show the frequency of occurrence in 50% majority rule consensus tree and numbers below branches are bootstrap values >50% based on 1000 replicates. Branches without numbers had bootstrap values <50%. Numbers in brackets show branch lengths (number of nucleotide substitution).

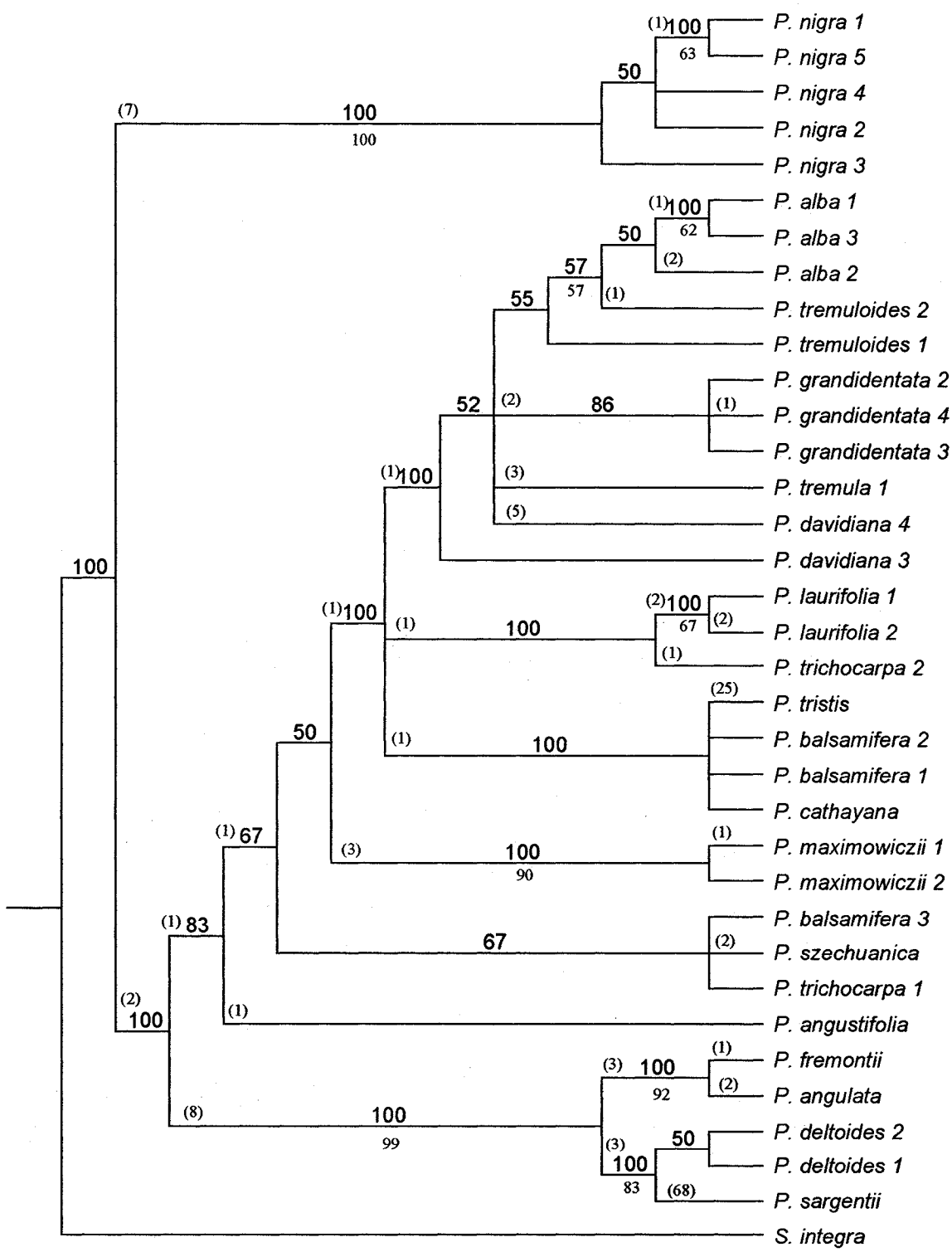

P. nigra 1
P. nigra 5
P. nigra 4
P. nigra 2
P. nigra 3
P. alba 1
P. alba 3
P. alba 2
P. tremuloides 2
P. tremuloides 1
P. grandidentata 2
P. grandidentata 4
P. grandidentata 3
P. tremula 1
P. davidiana 4
P. davidiana 3
P. laurifolia 1
P. laurifolia 2
P. trichocarpa 2
P. tristis
P. balsamifera 2
P. balsamifera 1
P. cathayana
P. maximowiczii 1
P. maximowiczii 2
P. balsamifera 3
P. szechuanica
P. trichocarpa 1
P. angustifolia
P. fremontii
P. angulata
P. deltoides 2
P. deltoides 1
P. sargentii
S. integra

Figure 2.7 The 50% majority rule consensus tree of 660 equally parsimonious trees (tree length 1553; CI = 0.800) based on combined six SCISSR fragments (PIS3A2, PIS5X1B4, PIS5A1, PIS6A2, PIS8A2, and PIS9A6) from Populus species. Numbers above branches show the frequency of occurrence in 50% majority rule consensus tree and numbers below branches are bootstrap values >50% based on 1000 replicates. Branches without numbers had bootstrap values <50%. Numbers in brackets show branch lengths (number of nucleotide substitution). Section affiliations denoted as A (Aigeiros) P (Populus), T (Tacamahaca) and geographic origins of species indicated as Asi (Asia), Nam (North America), Eus (Eurasia) are placed only next to the first individual of a taxon.

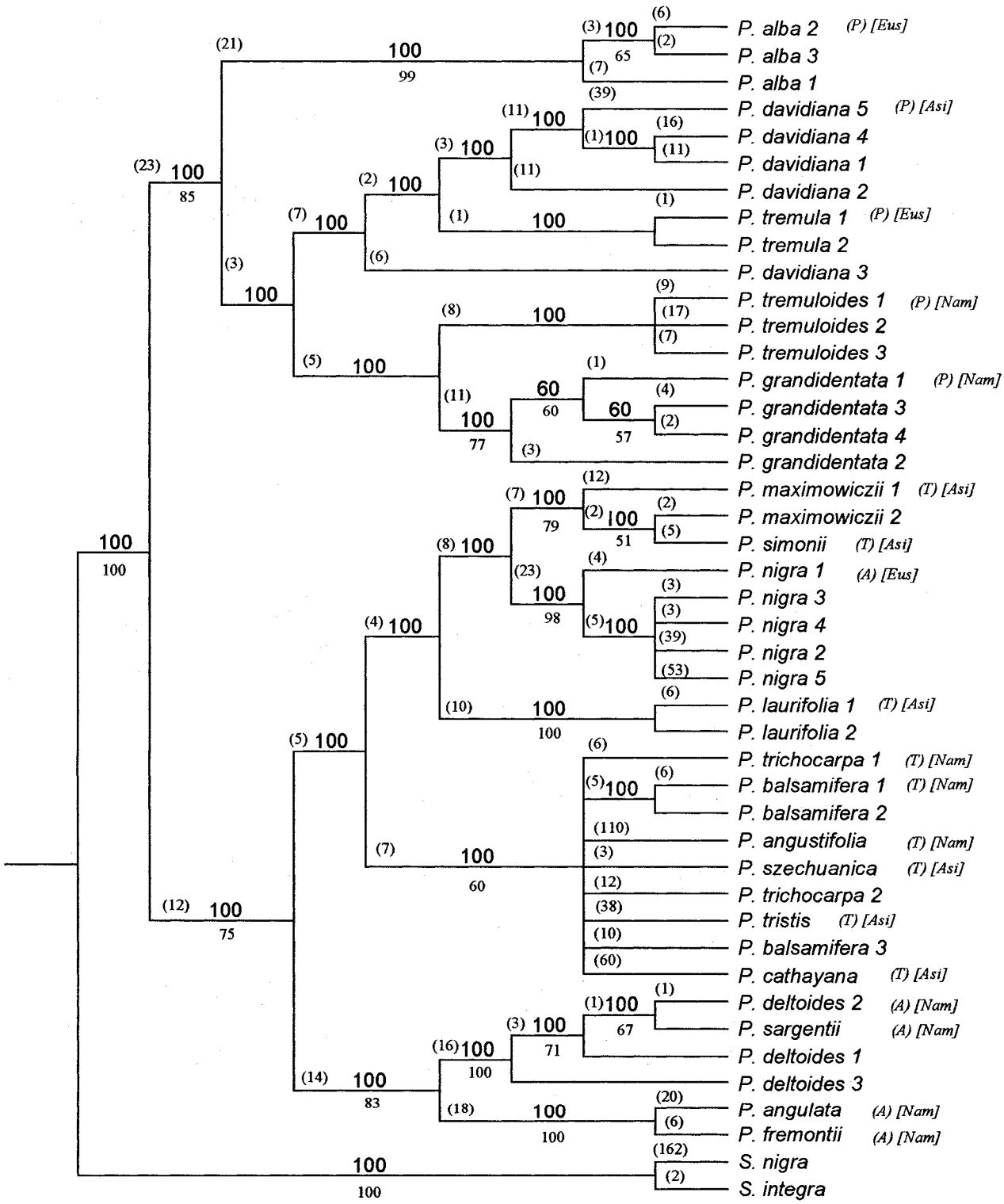
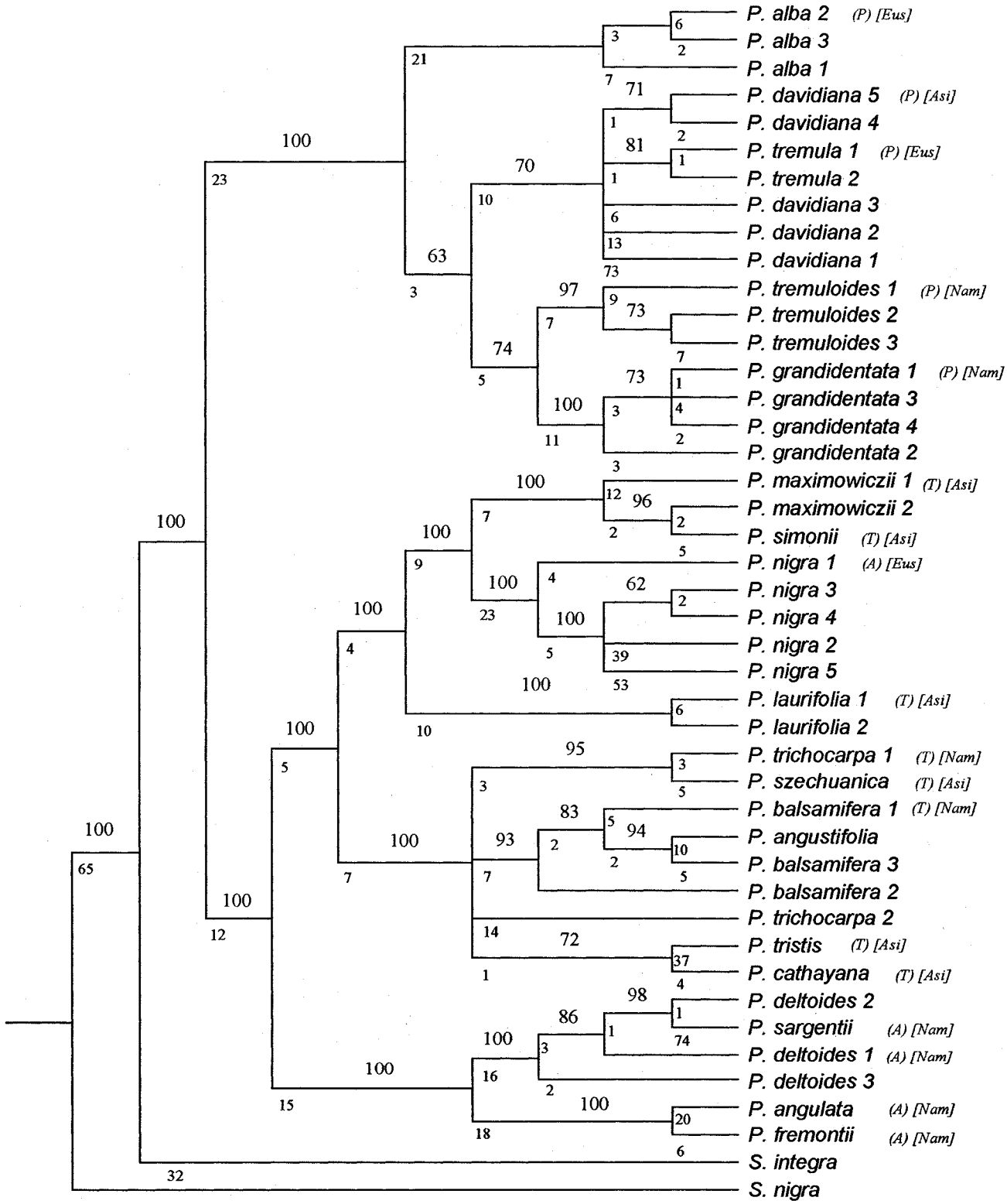

P. alba 2 (P) [Eus]
P. alba 3
P. alba 1
P. davidiana 5 (P) [Asi]
P. davidiana 4
P. davidiana 1
P. davidiana 2
P. tremula 1 (P) [Eus]
P. tremula 2
P. davidiana 3
P. tremuloides 1 (P) [Nam]
P. tremuloides 2
P. tremuloides 3
P. grandidentata 1 (P) [Nam]
P. grandidentata 3
P. grandidentata 4
P. grandidentata 2
P. maximowiczii 1 (T) [Asi]
P. maximowiczii 2
P. simonii (T) [Asi]
P. nigra 1 (A) [Eus]
P. nigra 3
P. nigra 4
P. nigra 2
P. nigra 5
P. laurifolia 1 (T) [Asi]
P. laurifolia 2
P. trichocarpa 1 (T) [Nam]
P. balsamifera 1 (T) [Nam]
P. balsamifera 2
P. angustifolia (T) [Nam]
P. szechuanica (T) [Asi]
P. trichocarpa 2
P. tristis (T) [Asi]
P. balsamifera 3
P. cathayana (T) [Asi]
P. deltoides 2 (A) [Nam]
P. sargentii (A) [Nam]
P. deltoides 1
P. deltoides 3
P. angulata (A) [Nam]
P. fremontii (A) [Nam]
S. nigra
S. integra

Figure 2. 8 Bayesian consensus of 9001 trees derived from the analyses of combined six SCISSR fragments (PIS3A2, PIS5X1B4, PIS5A1, PIS6A2, PIS8A2, and PIS9A6) from *Populus* species. Bayesian posterior probability values greater than 50% are shown above branches and numbers below branches show branch lengths (number of nucleotide substitution). Section affiliations denoted as A (*Aigeiros*), P (*Populus*), T (*Tacamahaca*) and geographic origins of species indicated as Asi (Asia), Nam (North America), Eus (Eurasia) are placed only next to the first individual of a taxon.


P. alba 2 (P) [Eus]
P. alba 3
P. alba 1
P. davidiana 5 (P) [Asi]
P. davidiana 4
P. tremula 1 (P) [Eus]
P. tremula 2
P. davidiana 3
P. davidiana 2
P. davidiana 1
P. tremuloides 1 (P) [Nam]
P. tremuloides 2
P. tremuloides 3
P. grandidentata 1 (P) [Nam]
P. grandidentata 3
P. grandidentata 4
P. grandidentata 2
P. maximowiczii 1 (T) [Asi]
P. maximowiczii 2
P. simonii (T) [Asi]
P. nigra 1 (A) [Eus]
P. nigra 3
P. nigra 4
P. nigra 2
P. nigra 5
P. laurifolia 1 (T) [Asi]
P. laurifolia 2
P. trichocarpa 1 (T) [Nam]
P. szechuanica (T) [Asi]
P. balsamifera 1 (T) [Nam]
P. angustifolia
P. balsamifera 3
P. balsamifera 2
P. trichocarpa 2
P. tristis (T) [Asi]
P. cathayana (T) [Asi]
P. deltoides 2
P. sargentii (A) [Nam]
P. deltoides 1 (A) [Nam]
P. deltoides 3
P. angulata (A) [Nam]
P. fremontii (A) [Nam]
S. integra
S. nigra

Appendix 2. 1 Nucleotide sequence data matrix of SCISSR fragment PIS3A2.

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[                          10         20         30         40         50]
[                          .          .          .          .          .]

P._alba_2                CAATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [49]
P._alba_3                CAATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [49]
P._alba_1                CAATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [49]
P._davidiana_5           CAATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCTACATGTATTGG    [49]
P._maximowiczii_1        --ATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [47]
P._laurifolia_1          --------------------AATATGAAAGGACAAAGCAACATGTATTGG    [30]
P._laurifolia_2          CAATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [49]
P._maximowiczii_2        CAATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [49]
P._nigra_1               CAATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [49]
P._simonii               ---TTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [46]
P._trichocarpa_1         CAATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [49]
P._balsamifera_1         ---TTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [46]
P._angustifolia          CAATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [49]
P._szechuanica           ----------------------TATGAAAGGACAAAGCAACATGTATTGG    [28]
P._trichocarpa_2         --ATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [47]
P._balsamifera_2         -AATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [48]
P._tristis               CAATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [49]
P._balsamifera_3         ---------TGACA-TGGGAAATATAAAAGGACAAAGCAACATGTATTGG    [40]
P._cathayana             --ATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [47]
P._nigra_3               AAATTACTTTGACA-TGGGGAATATGAAAGGACAAAGCAACATGTATTGG    [49]
P._nigra_4               --ATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [47]
P._nigra_2               -AATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [48]
P._deltoides_2           -AATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [48]
P._nigra_5               CAATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACTTGTTTGGC    [49]
P._angulata              CAATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACTTGTATTGG    [49]
P._fremontii             CAATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [49]
P._sargentii             CAATTACTTTGACAGTGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [50]
P._deltoides_1           CAATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [49]
P._deltoides_3           CAATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [49]
S._nigra                 ----------------------ATATGAAAGGACATAGCAACATGTATTGG    [29]
P._tremuloides_1         --ATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [47]
P._tremuloides_2         CAATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [49]
P._tremula_1             ---TTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [46]
P._grandidentata_1       ----------------------ATGAAAGGACAAAGCAACATGTATTGG    [27]
P._grandidentata_2       CAATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [49]
P._grandidentata_3       CAATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [49]
P._grandidentata_4       CAATTACTTTGACA-TGGGAAATATGAAAGGACAAAGCAACATGTATTGG    [49]
P._davidiana_3           -AATTACTTTGAC----------ATGAAGGGACAAAGCAACATGTATTGG    [39]
S._integra               ------------------------------------------------    [0]

[                          60         70         80         90        100]
[                          .          .          .          .          .]

P._alba_2                CACCAAAATCCGGCATTACTAGATGATAATGAACCTCAAAAATATTACTA    [99]
P._alba_3                CACCAAAATCCGGCATTACTAGATGATAATGAACCTCAAAAATATTACTA    [99]
P._alba_1                CACCAAAATCCGGCATTACTAGATGATAATGAACCTCAAAAATATTACTA    [99]
P._davidiana_5           CACCAAAATCCGGCATTACTAGATGATAATGAACCTCAAAAATATTACTA    [99]
P._maximowiczii_1        CACCATAATCTGGCATTTCTAGATGATAATGAACCTCAAA-ATATTACTA    [96]
P._laurifolia_1          CACCATAATCTGGCATTTCTAGATGATAATGAACCTCAGA-ATATTACTA    [79]
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P._laurifolia_2          CACCATAATCTGGCATTTCTAGATGATAATGAACCTCAAA-ATATTACTA    [98]
P._maximowiczii_2        CACCATAATCTGGCATTTCTAGATGATAATGAACCTCAAA-ATATTACTA    [98]
P._nigra_1               CACCATAATCCGGCATTTCTAGATGATAATGAACCTCAAA-ATATCACTA    [98]
P._simonii               CACCATAATCTGGCATTTCTAGATGATAATGAACCTCAAA-ATATTACTA    [95]
P._trichocarpa_1         CACCATAATCTGGCATTTCTAGATGATAATGAACCTCAAA-ATATTACTA    [98]
P._balsamifera_1         CACCATAATCTGGCATTTCTAGATGATAATGAACCTCAAA-ATATTACTA    [95]
P._angustifolia          CACCATAATCTGGCATTTCTAGATGATAATGAACCTCAAA-ATATTACTA    [98]
P._szechuanica           CACCATAATCTGGCATTTCTAGATGATAATGAACCTCAAA-ATATTACTA    [77]
P._trichocarpa_2         CACCATAATCTGGCATTTCTAGATGATAATGAACCTCAAA-ATATTACTA    [96]
P._balsamifera_2         CACCATAATCTGGCATTTCTAGATGATAATGAACCTCAAA-ATATTACTA    [97]
P._tristis               CACCATAATCTGGCATTTCTAGATGATAATGAACCTCAAA-ATATTACTA    [98]
P._balsamifera_3         CACCATAATCTGGCATTTCTAGATGATAATGAACCTCAAA-ATATTACTA    [89]
P._cathayana             CACCATAATCTGGCATTTCTAGATGATAATGAACCTCAGA-ATATTACTA    [96]
P._nigra_3               CACCATAATCCGGCATTTCTAGATGATAATGAACCTCAAA-ATATTACTA    [98]
P._nigra_4               CACCATAATCCGGCATTTCTAGATGATAATGAACCTCAAA-ATATTACTA    [96]
P._nigra_2               CACCATAATCCGGCATTTCTAGATGATAATGAACCTCAAA-ATATTACTA    [97]
P._deltoides_2           CACCATAATCCGGCATTTCTAGATGATAATGAACCTCAAA-ATATTACTA    [97]
P._nigra_5               CTGCGTAATCTGGCATGACTAGATGATAATGAACCTCAGA-ATATTACTA    [98]
P._angulata              CACCATAATCCGGCATTTCTAGATGATAATGAACCTCACA-ATATTACTG    [98]
P._fremontii             CACCATAATCCGGCATTTCTAGATGATAATGAACCTCACA-ATATTACTA    [98]
P._sargentii             CACCATAATCCGGCATTTCTAGATGATAATGAACCTCAAA-ATATTACTA    [99]
P._deltoides_1           CACCATAATCCGGCATTTCTAGATGATAATGAACCTCAAA-ATATTACTA    [98]
P._deltoides_3           CACCATAATCCGGCATTTCTAGATGATAATGAACCTCAAA-ATATTACTA    [98]
S._nigra                 CTCCAA-ATCTGGCATTAATAGATGGTTATGAACCTCAAA-ATATTGCTA    [77]
P._tremuloides_1         CACCAAAATCCGGCATTACTAGATGATAATGAACCTCAAA-ATATTACTA    [96]
P._tremuloides_2         CACCAAAATCCGGCATTACTAGATGATAATGAACCTCAAA-ATATTACTA    [98]
P._tremula_1             CACCAAAATCCGGCATTACTAGATGATAATGAACCTCAAA-ATATTACTA    [95]
P._grandidentata_1       CACCAAAATCCGGCATTACTAGATGATAATGAACCTCAAA-ATATTACTA    [76]
P._grandidentata_2       CACCAAAATCCGGCATTACTAGATGATAATGAACCTCAAA-ATATTACTA    [98]
P._grandidentata_3       CACCAAAATCCGGCATTACTAGATGATAATGAACCTCAAA-ATATTACTA    [98]
P._grandidentata_4       CACCAAAATCCGGCATTACTAGATGATAATGAACCTCAAA-ATATTACTA    [98]
P._davidiana_3           CACCAAAATCCGGCATTACTAGATGATAATGAACCTCAAA-ATATTACTA    [88]
S._integra               --------------------------------------------------    [0]

[                                110       120       130       140       150]
[                                .         .         .         .         .]

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P._alba_1                AGATTTTCAGTCACAGGGAAGGGAAGCCAATTATAGGGATAAATGTGGG-    [148]
P._davidiana_5           AGATTTTCAGTCACAGGGAAGGGAAGCCAATTATAGGGATAAGTGTGGG-    [148]
P._maximowiczii_1        AGATTTTCAGTCACAGGGAAGGGAAGCCAAATATAGGGATAATTGTGGG-    [145]
P._laurifolia_1          AGATTTTCAGTCACAGGGAAGGGAAGCCAAATATAGGGATAATTGTGGG-    [128]
P._laurifolia_2          AGATTTTCAGTCACAGGGAAGGGAAGCCAAATATAGGGATAATTGTGGG-    [147]
P._maximowiczii_2        AGATTTTCAGTCACAGGGAAGGGAAGCCAAATATAGGGATAATTGTGGG-    [147]
P._nigra_1               AGATTTTCAGTCACAGGGAAGGGAAGCCAAATATAGGGATAATTGTGGG-    [147]
P._simonii               AGATTTTCAGTCACAGGGAAGGGAAGCCAAATATAGGGATAATTGTGGG-    [144]
P._trichocarpa_1         AGATTTTCAATCACAGGGAAGGGAAGCCAAATATAGGGATAATTGTGGG-    [147]
P._balsamifera_1         AGATTTTCAATCACAGGGAAGGGAAGCCAAATATAGGGATAATTGTGGG-    [144]
P._angustifolia          AGATTTTCAGTCACAGGGAAGGGAAGCCAAATATAGGGATAATTGTGGG-    [147]
P._szechuanica           AGATTTTCAGTCACAGGGAAGGGAAGCCAAATATAGGGATAATTGTGGG-    [126]
P._trichocarpa_2         AGATTTTCAGTCACAGGGAAGGGAAGCCAAATATAGGGATAATTGTGGG-    [145]
P._balsamifera_2         AGATTTTCAGTCACAGGGAAGGGAAGCCAAATATAGGGATAATTGTGGG-    [146]
P._tristis               AGATTTTCAGTCACAGGGAAGGGAAGCCAAATATAGGGATAATTGTGGG-    [147]
P._balsamifera_3         AGATTTTCAGTCACAGGGAAGGGAAGCCAAATATAGGGATAATTGTGGG-    [138]
P._cathayana             AGATTTTCAGTCACAGGGAAGGGAAGCCAAATATAGGGATAATTGTGGG-    [145]
P._nigra_3               AGATTTTCAGTCACAGGGAAGGGAAGCCAAATACAGGGATAATTGTGGG-    [147]
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P._nigra_4              AGATTTTCAGTCACAGGGAAGGGAAGCCAAATACAGGGATAATTGTGGG-    [145]
P._nigra_2              AGATTTTCAGTCACAGGGAAGGGAAGCCAAATACAGGGATAATTGTGGG-    [146]
P._deltoides_2          AGATTTTCAGTCACAGGGAAGGGAAGCCAAATACAGGGATAATTGTGGG-    [146]
P._nigra_5              AGATTCTCAGTCACAGGGAAGGGAAGCCAAATACAGGGATAATTGTGCGC    [148]
P._angulata             CGATTTTCAATCACAGGGAAGGGAAGACAAATACAGGGATAATTGTGGG-    [147]
P._fremontii            AGATTTTCAATCACAGGGAAGGGAAGCCAAATACAGGGATAATTGTGGG-    [147]
P._sargentii            AGATTTTCAGTCACAGGGAAGGGAAGCCAAATACAGGGATAATTGTGGG-    [148]
P._deltoides_1          AGATTTTCAGTCACAGGGAAGGGAAGCCAAATACAGGGATAATTGTGGG-    [147]
P._deltoides_3          AGATTTTCAGTCACAGGGAAGGGAAGACAAATACAGGGATAATTGTGGG-    [147]
S._nigra                AGATTTTCAGTCACAGGGAAGGATATCGAAATATAGGGATAACTGCGGG-    [126]
P._tremuloides_1        AGATTTTCAGTCACAGGGAAGGGAAGCCAATTATAGGGATAAGTGTGGG-    [145]
P._tremuloides_2        AGATTTTCAGTCACAGGGAAGGGAAGCCAATTATAGGGATAAGTGTGGG-    [147]
P._tremula_1            AGAATTTCAGTCACAGGGAAGGGAAGCCAAATATAGGGATAAGTGTGGC-    [144]
P._grandidentata_1      AGATTTTCAGTCACAGGGAAGGGAAGCCAATTATAGGGATAAGTGTGGG-    [125]
P._grandidentata_2      AGATTTTCAGTCACAGGGAAGGGAAGCCAATTATAGGGATAAGTGTGGG-    [147]
P._grandidentata_3      AGATTTTCAGTCACAGGGAAGGGAAGCCAATTATAGGGATAAGTGTGGG-    [147]
P._grandidentata_4      AGATTTTCAGTCACAGGGAAGGGAAGCCAATTATAGGGATAAGTGTGGG-    [147]
P._davidiana_3          AGATTTTCAGTCACAGGGAAGGGAAGCCAATTATAGGGATAAGTGTGGG-    [137]
S._integra              --------------------------------------------------    [0]

[                                160       170       180       190       200]
[                                .         .         .         .         .]

P._alba_2               ATCCA--TGTATTCGCACCAAAGTCGGGAATGAAAGCAT----------    [185]
P._alba_3               ATCCA--TGTATTCGCACCAAAGTCGGGAATGAAAGCAT----------    [185]
P._alba_1               ATCCA--TGTATTCGCACCAAAGTCGGGAATGAAAGCAT----------    [185]
P._davidiana_5          ATCCA--TGTATTCGCACCAAAGTCGGGAATGAAAGCAT----------    [185]
P._maximowiczii_1       ATCCA--TGTATTCGCACCAAAGTCAGGAATGAAACCAT----------    [182]
P._laurifolia_1         ATCCA--TGTATTCGCACCAAAGTCAGGAATGAAACCAT----------    [165]
P._laurifolia_2         ATCCA--TGTATTCGCACCAAAGTCAGGAATGAAACCAT----------    [184]
P._maximowiczii_2       ATCCA--TGTATTCGCACCAAATTCAGGAATGAAACCAT----------    [184]
P._nigra_1              ATCCA--TGTATTCGCACCAAAGCCAGGAATGAAACCAT----------    [184]
P._simonii              ATCCA--TGTATTCGCACCAAAGTCAGGAATGAAACCAT----------    [181]
P._trichocarpa_1        ATCCA--TGTATTCGCACCAAAGTCAGGAATGAAACCAT----------    [184]
P._balsamifera_1        ATCCA--TGTATTCGCACCAAAGTCAGGAATGAAACCAT----------    [181]
P._angustifolia         ATCCA--TGTATTCGCACCAAAGTCAGGAGTGAAACCAT----------    [184]
P._szechuanica          ATCCA--TGTATTCGCACCAAAGTCAGGAGTGAAACCAT----------    [163]
P._trichocarpa_2        ATCCA--TGTATTCGCACCAAAGTCAGGAATGAAACCAT----------    [182]
P._balsamifera_2        ATCCA--TGTATTCGCACCAAAGTCAGGAATGAAACCAT----------    [183]
P._tristis              ATCCA--TGTATTCGCACCAAAGTCAGGAATGAAACCAT----------    [184]
P._balsamifera_3        ATCCA--TGTATTCGCACCAAAGTCAGGAATGAAACCAT----------    [175]
P._cathayana            ATCCA--TGTATTCGCACCAAAGTCAGGAATGAAACCATGAATGAAACCA   [193]
P._nigra_3              ATCCA--TGTATTCGCACCAAAGTCAGGAATGAAAACAT----------    [184]
P._nigra_4              ATCCA--TGTATTCGCACCAAAGTCAGGAATGAAAACAT----------    [182]
P._nigra_2              ATCCA--TGTATTCGCACCAAAGTCAGGAATGAAAACAT----------    [183]
P._deltoides_2          ATCCA--TGTATTCGCACCAAAGTCAGGAATGAAAACAT----------    [183]
P._nigra_5              ATCCA--TGTGTTCGCACCATAGTCGGGAATGAGAGCAT----------    [185]
P._angulata             ATCCA--TGTGTTCGCACCATAGTCAGGAATGAAACCGT----------    [184]
P._fremontii            ATCCA--TGTGTTCGCACCAAAGTCGGGAATGAAAACAT----------    [184]
P._sargentii            ATCCA--TGTATTCGCACCAAAGTCAGGAATGAAAACAT----------    [185]
P._deltoides_1          ATCCA--TGTATTCGCACCAAAGTCAGGAATGAAAACAT----------    [184]
P._deltoides_3          ATCCA--TGTATTCGCACCAAAGTCAGGAATGAAACCAT----------    [184]
S._nigra                ACTCCATTATATTCGCACCAGAATCAGGAATGAAACCAT----------    [165]
P._tremuloides_1        ATCCA--TGTATTCGCACCAAAGTCGGGAATGAAAGCAT----------    [182]
P._tremuloides_2        ATCCA--TGTATTCGCACCAAAGTCGGGAATGAAAGCAT----------    [184]
P._tremula_1            ATCCA--TGTATTCGCACCAAAGTCGGGAATGAAAGCAT----------    [181]
P._grandidentata_1      ATCCA--TGTATTCGCACCAAAGTCGGGAATGAAAGCAT----------    [162]
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P._grandidentata_2    ATCCA--TGTATTCGCACCAAAGTCGGGAATGAAAGCAT-----------    [184]
P._grandidentata_3    ATCCA--TGTATTCGCACCAAAGTCGGGAATGAAAGCAT-----------    [184]
P._grandidentata_4    ATCCA--TGTATTCGCACCAAAGTCGGGAATGAAAGCAT-----------    [184]
P._davidiana_3        ATCCA--TGTATTCGCACCAAAGTCGGGAATGAAAGCAT-----------    [174]
S._integra            --------------------------------------------------    [0]

[                              210        220        230        240        250]
[                              .          .          .          .          .]

P._alba_2             -AAAACAGCCAAAGCATTACAATGATGGTGAGTGGACATTACTTGGGGCA    [234]
P._alba_3             -AAAACAGCCAAAGCATTACAATGATGGTGAGTGGACATTACTTGGGGCA    [234]
P._alba_1             -AAAACAGCCAAAGCATTACAATGATGGTGAGTGGACATTACTTGGGGCA    [234]
P._davidiana_5        -AAAACAGCCAAAGCATTACAATGATGGTGAGTGGAAATTACTTGGGGCA    [234]
P._maximowiczii_1     -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAATTACTTGGGGCA    [231]
P._laurifolia_1       -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAATTACATGGGGCA    [214]
P._laurifolia_2       -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAATTACATGGGGCA    [233]
P._maximowiczii_2     -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAATTACTTGGGGCA    [233]
P._nigra_1            -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAATTACTTGGGGCA    [233]
P._simonii            -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAATTACTTGGGGCA    [230]
P._trichocarpa_1      -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAATTACTTGGGGCA    [233]
P._balsamifera_1      -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAATTACTTGGGGCA    [230]
P._angustifolia       -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAATTACTTGGGGCA    [233]
P._szechuanica        -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAATTACTTGGGGCA    [212]
P._trichocarpa_2      -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAATTACTTGGGGCA    [231]
P._balsamifera_2      -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAATTACTTGGGGCA    [232]
P._tristis            -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAATTACTTGGGGCA    [233]
P._balsamifera_3      -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAATTACTTGGGGCA    [224]
P._cathayana          TAAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAATTACTTGGGGCA    [243]
P._nigra_3            -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAATTACTTGGGGCA    [233]
P._nigra_4            -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAATTACTTGGGGCA    [231]
P._nigra_2            -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAATTACTTGGGGCA    [232]
P._deltoides_2        -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAGTTACTTGGGGCA    [232]
P._nigra_5            -AAAACAGCCCAAGCATTAAAATGATGGTGAGTGCAAATTACTTGGGGCA    [234]
P._angulata           -AAAACATCCTAAGCATTAAAATGATAGTGAGTGGAAATTACTTGGGGCA    [233]
P._fremontii          -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAGTTACTTGGGGCA    [233]
P._sargentii          -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAGTTACTTGGGGCA    [234]
P._deltoides_1        -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAGTTACTTGGGGCA    [233]
P._deltoides_3        -AAAACAGCCAAAGCATTAAAATGATGGTGAGTGGAAATTACTTGGGGCA    [233]
S._nigra              -AAAACATCCAAAGCATTTGAATGATGGTGGGTGGAACTTACTTGGGGCA    [214]
P._tremuloides_1      -AAAACAGCCAAAGCATTACAATGATGGTGAGTGGAAATTACTTGGGGCA    [231]
P._tremuloides_2      -AAAACAGCCAAAGCATTACAATGATGGTGAGTGGAAATTACTTGGGGCA    [233]
P._tremula_1          -AAAACAGCCAAAGCATTACAATGATGGTGTTTGGAAATTACTTGGGGCA    [230]
P._grandidentata_1    -AAAACAGCCAAAGCATTACAATGATGGTGAGTGGAAATTACTTGGGGCA    [211]
P._grandidentata_2    -AAAACAGCCAAAGCATTACAATGATGGTGAGTGGAAATTACTTGGGACA    [233]
P._grandidentata_3    -AAAACAGCCAAAGCATTACAATGATGGTGAGTGGAAATTACTTGGGGCA    [233]
P._grandidentata_4    -AAAACAGCCAAAGCATTACAATGATGGTGAGTGGAAATTACTTGGGGCA    [233]
P._davidiana_3        -AAAACAGCCAAAGCATTACAATGATGGTGAGTGGAAATTACTTGGGGCA    [223]
S._integra            --------------------------------------------------    [0]

[                              260        270        280        290        300]
[                              .          .          .          .          .]

P._alba_2             GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTGAAAGTG    [284]
P._alba_3             GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTGAAAGTG    [284]
P._alba_1             GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTGAAAGTG    [284]
P._davidiana_5        GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGTG    [284]
P._maximowiczii_1     GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG    [281]
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P._laurifolia_1        GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [264]
P._laurifolia_2        GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [283]
P._maximowiczii_2      GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [283]
P._nigra_1             GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [283]
P._simonii             GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [280]
P._trichocarpa_1       GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [283]
P._balsamifera_1       GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [280]
P._angustifolia        GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [283]
P._szechuanica         GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [262]
P._trichocarpa_2       GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [281]
P._balsamifera_2       GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [282]
P._tristis             GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [283]
P._balsamifera_3       GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [274]
P._cathayana           GATGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [293]
P._nigra_3             GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [283]
P._nigra_4             GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [281]
P._nigra_2             GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [282]
P._deltoides_2         GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [282]
P._nigra_5             CAGGAAGCTGCACAAGAGTGCCATGAATATATGGGTCCTCGTTCAAAGCG   [284]
P._angulata            CAGGAAGCTGCACATGAATACCATGAATATATGGGTCCTCGTTAAAAGCG   [283]
P._fremontii           GAGGAAGCTGCACAAGAATACCATGAATATATGGGTCCTCGTTAAAAGCG   [283]
P._sargentii           GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [284]
P._deltoides_1         GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [283]
P._deltoides_3         GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [283]
S._nigra               GAGGAAGCTGCACAAGAAGACCATGAATCGATGGGTCCTCGTTAAAAGTG   [264]
P._tremuloides_1       GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [281]
P._tremuloides_2       GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGCG   [283]
P._tremula_1           GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTGAAAGCG   [280]
P._grandidentata_1     GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGTG   [261]
P._grandidentata_2     GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGTG   [283]
P._grandidentata_3     GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGTG   [283]
P._grandidentata_4     GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGTG   [283]
P._davidiana_3         GAGGAAGCTGCACAAGAATACCATGAATAGATGGGTCCTCGTTAAAAGTG   [273]
S._integra             --------------------------------------------------   [0]

[                              310        320        330        340        350]
[                              .          .          .          .          .]

P._alba_2              GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGGGGGAACTGTGT   [334]
P._alba_3              GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGGGGGAACTGTGT   [334]
P._alba_1              GACAAGGCATGAATAACAACATCTTGTTTACAGTCTTGGGGGAACTGTGT   [334]
P._davidiana_5         GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGGGGGAACTGTGT   [334]
P._maximowiczii_1      GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [331]
P._laurifolia_1        GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [314]
P._laurifolia_2        GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [333]
P._maximowiczii_2      GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [333]
P._nigra_1             GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [333]
P._simonii             GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [330]
P._trichocarpa_1       GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [333]
P._balsamifera_1       GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [330]
P._angustifolia        GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [333]
P._szechuanica         GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [312]
P._trichocarpa_2       GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [331]
P._balsamifera_2       GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [332]
P._tristis             GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [333]
P._balsamifera_3       GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [324]
P._cathayana           GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [343]
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P._nigra_3              GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [333]
P._nigra_4              GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [331]
P._nigra_2              GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [332]
P._deltoides_2          GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [332]
P._nigra_5              GACAAGGCATGAATAACCTCATCTTGTTTACAATCTTGCGGGAACTGTGT   [334]
P._angulata             GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGA   [333]
P._fremontii            GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [333]
P._sargentii            GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [334]
P._deltoides_1          GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGT   [333]
P._deltoides_3          GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGCGGGAACTGTGA   [333]
S._nigra                GACAAAGCACTAATAACATCATCTTGATTGCAATTTTGGGGGAACTGTGT   [314]
P._tremuloides_1        GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGGGGGAACTGTGT   [331]
P._tremuloides_2        GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGGGGGAACTGTGT   [333]
P._tremula_1            GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGGGGGAACTGTGT   [330]
P._grandidentata_1      GACACGGCATGAATAACATCATCGTGTTTACAATCTTGGGGGAACTGTGT   [311]
P._grandidentata_2      GACAAGGCATGAATAACATCATCGTGTTTACAATCTTGGGGGAACTGTGT   [333]
P._grandidentata_3      GACAAGGCATGAATAACATCATCGTGTTTACAATCTTGGGGGAACTGTGT   [333]
P._grandidentata_4      GACAAGGCATGAATAACATCATCGTGTTTACAATCTTGGGGGAACTGTGT   [333]
P._davidiana_3          GACAAGGCATGAATAACATCATCTTGTTTACAATCTTGGGGGAACTGTGT   [323]
S._integra              ----------------CATCATCTTGATTGCGATTTTGGGGGAACAGTGT   [34]

[                                360       370       380       390       400]
[                                .         .         .         .         .]

P._alba_2               GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [384]
P._alba_3               GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [384]
P._alba_1               GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [384]
P._davidiana_5          GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [384]
P._maximowiczii_1       GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [381]
P._laurifolia_1         GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [364]
P._laurifolia_2         GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [383]
P._maximowiczii_2       GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [383]
P._nigra_1              GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [383]
P._simonii              GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [380]
P._trichocarpa_1        GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [383]
P._balsamifera_1        GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [380]
P._angustifolia         GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [383]
P._szechuanica          GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [362]
P._trichocarpa_2        GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [381]
P._balsamifera_2        GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [382]
P._tristis              GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [383]
P._balsamifera_3        GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [374]
P._cathayana            GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [393]
P._nigra_3              GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [383]
P._nigra_4              GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [381]
P._nigra_2              GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [382]
P._deltoides_2          GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [382]
P._nigra_5              GACATGAGACTTGATTCCAGCATCCTCGCAATGTTTAACCTTGTTGCGAA   [384]
P._angulata             GACAACAGACTTGTTTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [383]
P._fremontii            GACAACAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [383]
P._sargentii            GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [384]
P._deltoides_1          GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [383]
P._deltoides_3          GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [383]
S._nigra                GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTATAACCTTGTTGCGAA   [364]
P._tremuloides_1        GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [381]
P._tremuloides_2        GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [383]
P._tremula_1            GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [380]
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P._grandidentata_1    GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [361]
P._grandidentata_2    GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [383]
P._grandidentata_3    GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [383]
P._grandidentata_4    GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [383]
P._davidiana_3        GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTTTAACCTTGTTGCGAA   [373]
S._integra            GACAAGAGACTTGATTCCAGCATCCTCGCAAGCTATAACCTTGTTGCGAA   [84]

[                              410        420        430        440        450]
[                              .          .          .          .          .]

P._alba_2             CGTATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [434]
P._alba_3             CGTATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [434]
P._alba_1             CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [434]
P._davidiana_5        CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [434]
P._maximowiczii_1     CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [431]
P._laurifolia_1       CATATGTAAGAGAGTCCCTTCTGTCGCCCACAAGAATTACAGCCAAGCCA   [414]
P._laurifolia_2       CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [433]
P._maximowiczii_2     CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [433]
P._nigra_1            CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [433]
P._simonii            CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [430]
P._trichocarpa_1      CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [433]
P._balsamifera_1      CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [430]
P._angustifolia       CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [433]
P._szechuanica        CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [412]
P._trichocarpa_2      CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [431]
P._balsamifera_2      CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [432]
P._tristis            CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [433]
P._balsamifera_3      CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [424]
P._cathayana          CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCTAGCCA   [443]
P._nigra_3            CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [433]
P._nigra_4            CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [431]
P._nigra_2            CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [432]
P._deltoides_2        CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [432]
P._nigra_5            CATATGTAAGAGAGTCCCTTCTGTCGCCCTCGAGAATTACAGCCAGACCG   [434]
P._angulata           CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAACCG   [433]
P._fremontii          CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAACCA   [433]
P._sargentii          CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [434]
P._deltoides_1        CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [433]
P._deltoides_3        CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [433]
S._nigra              CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATCACAGCCAAGCCA   [414]
P._tremuloides_1      CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAATCCA   [431]
P._tremuloides_2      CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAATCCA   [433]
P._tremula_1          CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAATCCA   [430]
P._grandidentata_1    CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [411]
P._grandidentata_2    CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [433]
P._grandidentata_3    CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [433]
P._grandidentata_4    CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [433]
P._davidiana_3        CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATTACAGCCAAGCCA   [423]
S._integra            CATATGTAAGAGAGTCCCTTCGGTCGCCCACAAGAATCACAGCCAAGCCA   [134]

[                              460        470        480        490        500]
[                              .          .          .          .          .]

P._alba_2             GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCAGCAATTCT   [484]
P._alba_3             GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCAGCAATTCT   [484]
P._alba_1             GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCAGCAATTCT   [484]
P._davidiana_5        GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCAGCAATTCT   [484]
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P._maximowiczii_1     GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACCTGTTCAGCAATTCT   [481]
P._laurifolia_1       GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACCTGTTCAGCAATTCT   [464]
P._laurifolia_2       GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACCTGTTCAGCAATTCT   [483]
P._maximowiczii_2     GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACCTGTTCAGCAATTCT   [483]
P._nigra_1            GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACCTGTTCAGCAATTCT   [483]
P._simonii            GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACCTGTTCAGCAATTCT   [480]
P._trichocarpa_1      GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACCTGTTCAGCAATTCT   [483]
P._balsamifera_1      GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACCTGTTCAGCAATTCT   [480]
P._angustifolia       GGAATTTTTCCAACGGAATCCTTCATCCTCCTAACCTGTTCAGCAATTCT   [483]
P._szechuanica        GGAATTTTTCCAACGGAATCCTTCATCCTCCTAACCTGTTCAGCAATTCT   [462]
P._trichocarpa_2      GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACCTGTTCAGCAATTCT   [481]
P._balsamifera_2      GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACCTGTTCAGCAATTCT   [482]
P._tristis            GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACCTGTTCAGCAATTCT   [483]
P._balsamifera_3      GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACCTGTTCAGCAATTCT   [474]
P._cathayana          GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACCTGTTCAGCAATTCT   [493]
P._nigra_3            GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCAGCAATTCT   [483]
P._nigra_4            GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCAGCAATTCT   [481]
P._nigra_2            GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCAGCAATTCT   [482]
P._deltoides_2        GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCAGCAATTCT   [482]
P._nigra_5            GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCTTTAATTCT   [484]
P._angulata           TGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCATCAATTCT   [483]
P._fremontii          TGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCATCAATTCT   [483]
P._sargentii          GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCAGCAATTCT   [484]
P._deltoides_1        GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCAGCAATTCT   [483]
P._deltoides_3        GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCAGCAATTCT   [483]
S._nigra              GGAACTTTCCCAACGGAATCCTTCATCCTCCTAACTTGTTCGGCAATTCT   [464]
P._tremuloides_1      GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCAGCAATTCT   [481]
P._tremuloides_2      GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCAGCAATTCT   [483]
P._tremula_1          GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCAGCAATTCT   [480]
P._grandidentata_1    GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCAGCAATTCT   [461]
P._grandidentata_2    GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCAGCAATTCT   [483]
P._grandidentata_3    GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCAGCAATTCT   [483]
P._grandidentata_4    GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCAGCAATTCT   [483]
P._davidiana_3        GGAACTTTTCCAACGGAATCCTTCATCCTCCTAACTTGTTCAGCAATTCT   [473]
S._integra            GGAACTTTCCCAACTGAATCCTTCATCCTCCTAACTTGTTCGGCAATTCT   [184]

[                              510        520        530        540        550]
[                              .          .          .          .          .]

P._alba_2             GAACCTTATTTCATCCGCAATCAATTTGCCATCAATTACAAGAGCAGAAG   [534]
P._alba_3             GAACCTTATTTCATCCGCAATCAATTTGCCATCAATTACAAGAGCAGAAG   [534]
P._alba_1             GAACCTTATTTCATCCGCAATCAATTTTCCATCAATTACAAGAGCAGAAG   [534]
P._davidiana_5        GAACCTTATTTCATCCGCAATCAATTTTCCATCAATTACAAGAGCAGAAG   [534]
P._maximowiczii_1     GAACCTTATTTCATCCGCAATCAACTTGCCATCAATTACAAGAGCAGAAG   [531]
P._laurifolia_1       GAACCTTATTTCATCCGCAATCAACTTGCCATCAATTACAAGAGCAGAAG   [514]
P._laurifolia_2       GAACCTTATTTCATCCGCAATCAACTTGCCATCAATTACAAGAGCAGAAG   [533]
P._maximowiczii_2     GAACCTTATTTCATCCGCAATCAACTTGCCATCAATTACAAGAGCAGAAG   [533]
P._nigra_1            GAACCTTATTTCATCCGCAATCAACTTGCCATCAATTACAAGAGCAGAAG   [533]
P._simonii            GAACCTTATTTCATCCGCAATCAACTTGCCATCAATTACAAGAGCAGAAG   [530]
P._trichocarpa_1      GAACCTTATTTCATCCGCAATCAACTTGCCATCAATTACAAGAGCAGAAG   [533]
P._balsamifera_1      GAACCTTATTTCATCCGCAATCAACTTGCCATCAATTACAAGAGCAGAAG   [530]
P._angustifolia       GAACCTTATTTCATCCGCAATCAACTTGCCATCAATTACAAGAGCAGAAG   [533]
P._szechuanica        GAACCTTATTTCATCCGCAATCAACTTGCCATCAATTACAAGAGCAGAAG   [512]
P._trichocarpa_2      GAACCTTATTTCATCCGCAATCAACTTGCCATCAATTACAAGAGCAGAAG   [531]
P._balsamifera_2      GAACCTTATTTCATCCGCAATCAACTTGCCATCAATTACAAGAGCAGAAG   [532]
P._tristis            GAACCTTATTTCACCCGCAATCAACTTGCCATCAATTACAAGAGCAGAAG   [533]
P._balsamifera_3      GAACCTTATTTCATCCGCAATCAACTTGCCATCAATTACAAGAGCAGAAG   [524]
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P._cathayana          GAACCTTATTTCATCCGCAATCAACTTGCCATCAATTACAAGAGCAGAAG   [543]
P._nigra_3            GAACCTTATTTCATCCGCAATCATCTTGCCATCAATTACAAGAGCAGAAG   [533]
P._nigra_4            GAACCTTATTTCATCCGCAATCATCTTGCCATCAATTACAAGAGCAGAAG   [531]
P._nigra_2            GAACCTTATTTCATCCGCAATCATCTTGCCATCAATTACAAGAGCAGAAG   [532]
P._deltoides_2        GAACCTTATTTCATCCGCAATCATCTTGCCATCAATTACAAGAGCAGAAG   [532]
P._nigra_5            GAACCTTATTTCATCCGCAATCTGCTTGCCATCTCTTACAAGAATTTAAG   [534]
P._angulata           GAACCTTATTTCATCCGCAATCAACTTGCCATCTATTACAAGAGCATAAG   [533]
P._fremontii          GAACCTTATTTCATCCGCAATCATCTTGCCATCAATTACAAGAGCATAAG   [533]
P._sargentii          GAACCTTATTTCATCCGCAATCATCTTGCCATCAATTACAAGAGCAGAAG   [534]
P._deltoides_1        GAACCTTATTTCATCCGCAATCATCTTGCCATCAATTACAAGAGCAGAAG   [533]
P._deltoides_3        GAACCTTATTTCATCCGCAATCAACTTGCCATCAATTACAAGAGCAGAAG   [533]
S._nigra              GAACCTTATTTCATCCGCAATCAACTTGCCATCCATTACAAGAGCAACAG   [514]
P._tremuloides_1      GAACCTTATTTCATCCGCAATCAATTTTCCATCAATTACAAGAGCAAAAG   [531]
P._tremuloides_2      GAACCTTATTTCATCCGCAATCAATTTGCCATCAATTACAAGAGCAGAAG   [533]
P._tremula_1          GAACCTTATTTCATCCGCAATCAATTTGCCATCAATTACAAGAGCAGAAG   [530]
P._grandidentata_1    GAACCTTATTTCATCCGCAATCAATTTGCCATCAATTACAAGAGCAGAAG   [511]
P._grandidentata_2    GAACCTTATTTCATCCGCAATCAATTTGCCATCAATTACAAGAGCAGAAG   [533]
P._grandidentata_3    GAACCTTATTTCATCCGCAATCAATTTGCCATCAATTACAAGAGCAGAAG   [533]
P._grandidentata_4    GAACCTTATTTCATCCGCAATCAATTTGCCATCAATTACAAGAGCAGAAG   [533]
P._davidiana_3        GAACCTTATTTCATCCGCAATCAATTTGCCATCAATTACAAGAGCAGAAG   [523]
S._integra            GAACCTTATTTCATCCGCAATCAACTTGCCATCAATTACAAGAGCAGCAG   [234]

[                              560        570        580        590        600]
[                              .          .          .          .          .]

P._alba_2             AAGGTTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [583]
P._alba_3             AAGGTTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [583]
P._alba_1             AAGGTTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [583]
P._davidiana_5        AAGGTTGGTCATTAGTTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [583]
P._maximowiczii_1     AAGGGCGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [580]
P._laurifolia_1       AAGGGCGGTCATTATCTGTGCCTGTGTATTAAAAATAACAGAATTAATA-   [563]
P._laurifolia_2       AAGGGCGGTCATTAGCTGTGCCTGTGTATTAAAAATAACAGAATTAATA-   [582]
P._maximowiczii_2     AAGGGCGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [582]
P._nigra_1            AAGGGTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [582]
P._simonii            AAGGGTGGTCATTAGCAGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [579]
P._trichocarpa_1      AAGGGTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [582]
P._balsamifera_1      AAGGGTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [579]
P._angustifolia       AAGGGTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [582]
P._szechuanica        AAGGGTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [561]
P._trichocarpa_2      AAGGGTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [580]
P._balsamifera_2      AAGGGTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [581]
P._tristis            AAGGGTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [582]
P._balsamifera_3      AAGGGTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [573]
P._cathayana          AAGGGTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [592]
P._nigra_3            AAGGTTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [582]
P._nigra_4            AAGGTTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [580]
P._nigra_2            AAGGTTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [581]
P._deltoides_2        AAGGTTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [581]
P._nigra_5            ACGGTTGGTCATTATCTGCGCCTGTGTATTTAAAATAACGG---------   [575]
P._angulata           AAGGTTGGTCATTACCTGTGCCTGTGTATTTAAAATAACGGAAGTAATA-   [582]
P._fremontii          AAGGTTGGTCATTACCTGTGCCTGTGTATTTAAAATAACGGAATTAATA-   [582]
P._sargentii          AAGGTTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [583]
P._deltoides_1        AAGGTTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [582]
P._deltoides_3        AAGGTTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [582]
S._nigra              AAGGTTGGTCATTAGCTGTGCCTGCGTATTTAAAATAACGGAATTAATAA   [564]
P._tremuloides_1      AAGGCTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [580]
P._tremuloides_2      AAGGCTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [582]
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P._tremula_1          AAGGTTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [579]
P._grandidentata_1    AAGGTTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATT-   [560]
P._grandidentata_2    AAGGTTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATT-   [582]
P._grandidentata_3    AAGGTTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATT-   [582]
P._grandidentata_4    AAGGTTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATT-   [582]
P._davidiana_3        AAGGTTGGTCATTAGCTGTGCCTGTGTATTTAAAATAACAGAATTAATA-   [572]
S._integra            AAGGTTGGTCATTAGCTGTGCCTGCGTATTTAAAATAACGGAATTAATAA   [284]

[                              610       620       630       640       650]
[                              .         .         .         .         .]

P._alba_2             GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [632]
P._alba_3             GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [632]
P._alba_1             GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [632]
P._davidiana_5        GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [632]
P._maximowiczii_1     GATATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [629]
P._laurifolia_1       GAGATTGATTTTCTTTCACAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [613]
P._laurifolia_2       GAGATTTAATTTCTTTC-CAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [631]
P._maximowiczii_2     GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [631]
P._nigra_1            GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [631]
P._simonii            GATATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [628]
P._trichocarpa_1      GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [631]
P._balsamifera_1      GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [628]
P._angustifolia       GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [631]
P._szechuanica        GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [610]
P._trichocarpa_2      GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [629]
P._balsamifera_2      GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAA-TGA   [629]
P._tristis            GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAA-TGA   [630]
P._balsamifera_3      GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [622]
P._cathayana          GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [641]
P._nigra_3            GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [631]
P._nigra_4            GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [629]
P._nigra_2            GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [630]
P._deltoides_2        GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [630]
P._nigra_5            --------------------------------------------------   [575]
P._angulata           GACATTTCATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [631]
P._fremontii          GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [631]
P._sargentii          GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [632]
P._deltoides_1        GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [631]
P._deltoides_3        GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [631]
S._nigra              GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTAAAGGAAAATGA   [613]
P._tremuloides_1      GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [629]
P._tremuloides_2      GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [631]
P._tremula_1          GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTGGGGAAAATGA   [628]
P._grandidentata_1    GAGATTTAATGTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [609]
P._grandidentata_2    GAGATTTAATGTCTTTC-TGGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [631]
P._grandidentata_3    GAGATTTAATGTCTTTC-TGGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [631]
P._grandidentata_4    GAGATTTAATGTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [631]
P._davidiana_3        GAGATTTAATTTCTTTC-TAGCTAGTTATTGAAAAAGTTAGGGAAAATGA   [621]
S._integra            GAGATTTGATTTCTTTC-TAGCTACTTATTGAAAAAGTAAAGGAAAATGA   [333]

[                              660       670       680       690       700]
[                              .         .         .         .         .]

P._alba_2             CTAACAACAGTTCTTAATTGCTTGGGATGATGGAGGCCAAATGTCAGGGA   [682]
P._alba_3             CTAACAACAGTTCTTAATTGCTTGGGATGATGGAAGCCAAATGTCAGGGA   [682]
P._alba_1             CTAACAACAGTTCTTAATTGCTTGGGATGATGGAGGCCAAATGTCAGGGA   [682]
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P._davidiana_5          CTAACAACAGTTCTTAATTGCTTGGTATGATGGAGGCCAAATGTCAGGGA   [682]
P._maximowiczii_1       CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCAGGGA   [679]
P._laurifolia_1         CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCAGGGA   [663]
P._laurifolia_2         CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCAGGGA   [681]
P._maximowiczii_2       CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCAGGGA   [681]
P._nigra_1              CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCAGGGA   [681]
P._simonii              CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCAGGGA   [678]
P._trichocarpa_1        CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCAGGGA   [681]
P._balsamifera_1        CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCAGGGA   [678]
P._angustifolia         CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCAGGGA   [681]
P._szechuanica          CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCAGGGA   [660]
P._trichocarpa_2        CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCAGGGA   [679]
P._balsamifera_2        CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCAGGGA   [679]
P._tristis              CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCAGGGA   [680]
P._balsamifera_3        CTAACAACAGTTCTTAATTGCTGGGGATAATGGAGGCCAAATGTCAG---   [669]
P._cathayana            CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCAGGGA   [691]
P._nigra_3              CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCA----   [677]
P._nigra_4              CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCAGGGA   [679]
P._nigra_2              CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCAGGGA   [680]
P._deltoides_2          CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCAGGGA   [680]
P._nigra_5              --------------------------------------------------   [575]
P._angulata             CTAACAACTGTTCTTAATTGCTGGGGATGATGGAGGCCAAAT--------   [673]
P._fremontii            CTAACAACTGTTCTTAATTGCTGGGGATGATGGAGGCCAAATG-------   [674]
P._sargentii            CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCAGGGA   [682]
P._deltoides_1          CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCAGGGA   [681]
P._deltoides_3          CTAACAACAGTTCTTAATTGCTGGGGATGATGGAGGCCAAATGTCAGGGA   [681]
S._nigra                CTAAC-----------------------------------------------   [618]
P._tremuloides_1        CTAACAACAGTTCTTAATTGCTTGGGATGATGGAGGCCAAATGTCAGGGA   [679]
P._tremuloides_2        CTAACAACAGTTCTTAATTGCTTGGGATGATGGAGGCCAAATGTCAGGGA   [681]
P._tremula_1            CTAACAACAGTTCTTAATTGCTTGGGATGATGGAGGCCAAATGTCAGGGA   [678]
P._grandidentata_1      CTAACAACAGTTCTTAATTGCCTGGGATGATGGAGGCCAAATGTCAGGGA   [659]
P._grandidentata_2      CTAACAACAGTTCTTAATTGCCTGGGATGATGGAGGCCAAATGTCAGGGA   [681]
P._grandidentata_3      CTAACAACAGTTCTTAATTGCCTGGGATGATGGAGGCCAAATGTCAGGGA   [681]
P._grandidentata_4      CTAACAACAGTTCTTAATTGCCTGGGATGATGGAGGCCAAATGTCAGGGA   [681]
P._davidiana_3          CTAACAACAGTTCTTAATTGCTTGGGATGATGGAGGCCAAATGTCAGGGA   [671]
S._integra              CTAACAACAGTTAGTAACTGCCGGTGATGATGGAGGCCAAATGTCAGGGA   [383]

[                                710       720       730       740 ]
[                                .         .         .         .   ]

P._alba_2               GGTCAAGAGAGAGTAGAGATGAAGGAAACTTAAGAATTG----    [721]
P._alba_3               GGTCAAGAGAGAGTAGAGATGAAGGAAACTTAAGAATTG----    [721]
P._alba_1               GGTCAAGAGAGAGTAGAGATGAAGGAAACTTA-----------    [714]
P._davidiana_5          GGTCAAGAGAGAGTAGAGATGAAGGAAACTTA-----------    [714]
P._maximowiczii_1       GGTCAAGAGAGAGTAGAGATGAAGGAAACTTA-----------    [711]
P._laurifolia_1         GGTCAAGAGAGAGTAGAGATGAAGGAAACT-------------    [693]
P._laurifolia_2         GGTCAAGAGAGAGTAGAGATGAAGGAAACTTAAGAA-------    [717]
P._maximowiczii_2       GGTCAAGAGAGAGTAGAGATGAAGGA-----------------    [707]
P._nigra_1              GGTCGAGAGAGAGTAGAGATGAAGGAAACTTAAGA--------    [716]
P._simonii              GGTCAAGAGAGAGTAGAGATGAAGGAAACTTA-----------    [710]
P._trichocarpa_1        GATCAAGAGAGAGTAGAGATGAAGGAAACTTA-----------    [713]
P._balsamifera_1        GATCAAGAGAGAGTAGAGATGAAGGAAACTT------------    [709]
P._angustifolia         GATCAAGAGAGAGTAGAGATGAAGGAAACTTAAGAATTG----    [720]
P._szechuanica          GATCAAGAGAGAGTAGAGATGAAGGAAACTTA-----------    [692]
P._trichocarpa_2        GATCAAGAGAGAGTAGAGATGAAGGAAACTTAAGAATTG----    [718]
P._balsamifera_2        GATCAAGAGAGAGTAGAGATGAAGGAAACTTA-----------    [711]
P._tristis              GATCAAGAGAGAGTAGAGATGAAGGAAACTTAAG---------    [714]
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P._balsamifera_3      ----------------------------------------   [669]
P._cathayana          GATCAAGAGAGAGT-----------------------------   [705]
P._nigra_3            ----------------------------------------   [677]
P._nigra_4            GGTCAAGAGAGATTAAAGATGAAGGAAACTTAAGAATTGGGCT   [722]
P._nigra_2            GGTCAAGAGAGAGTAGAGATGAAGGAAACTTAAGAATTGGGCT   [723]
P._deltoides_2        GGTCAAGAGAGAGTAGAGATGAAGGAAACTTAAGAATTG----   [719]
P._nigra_5            ----------------------------------------   [575]
P._angulata           ----------------------------------------   [673]
P._fremontii          ----------------------------------------   [674]
P._sargentii          GGTCAAGAGAGAGTAGAGATGAAGGAAAC--------------   [711]
P._deltoides_1        GGTCAAGAGAGAGTAGAGATGAAGGAAACTTA-----------   [713]
P._deltoides_3        GGTCAAGAGAGAGTAGAGATGAAGGAAACTTAAG---------   [715]
S._nigra              ----------------------------------------   [618]
P._tremuloides_1      GGTC----------------------------------------   [683]
P._tremuloides_2      GGTCAAGAGAGAGTAGAGATGAAGGAAACTTAAGAATTGG---   [721]
P._tremula_1          GGTCAAGAGAGAGTAGAGACGAAGGAAACTTA-----------   [710]
P._grandidentata_1    GGTCAAGAGAGAGTAGAGATGAAGGAAACTTA-----------   [691]
P._grandidentata_2    GGTCAAGAGAGAGTAGAGATGAAGGAAACTTAAGAATTGGG--   [722]
P._grandidentata_3    GGTCAAGAGAGAGTAGAGATGAAGGAAACTTA-----------   [713]
P._grandidentata_4    GGTCAAGAGAGAGTAGAGATGAAGGAAACTTAAGAATTG----   [720]
P._davidiana_3        GGTCAAGAGAGAGTAGAGATGAAGGAAACTTAAGAATTGGGCT   [714]
S._integra            GGTCGAGGGAGAGTAGAGATGAGGGAAACTTAAGAATTGGGCT   [426]
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Appendix 2. 2 Nucleotide sequence data matrix of SCISSR fragment PIS5X1B4.

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[                          10         20         30         40         50]
[                          .          .          .          .          .]

P._alba_1                  ------------TGCCTTTTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [38]
P._alba_2                  CACTTCTTC-ATTGCCTTTTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [49]
P._alba_3                  --CTTCTTC-ATTGCCTTTTGTTTTTCTTCTTTGAACTTGACGCTCTGAC    [47]
P._tremula_1               CACTTCTTCC-ATGCCTTTTGTTTTTCTTCTTTCA-CTTGAAGCTCTGAC    [48]
P._tremula_2               CACTTCTTC-AATGCCTTTTGTTTTTCTTCTTTCA-CTTGAAGCTCTGAC    [48]
P._davidiana_5             CACTTCTTC-AATGCCTTTTGTTTTTCTTCTTTCA-CTTGAAGCTCTGAC    [48]
P._davidiana_4             CACTTCTTC-AATGCCTTTTGTTTTTCTTCTTTCA-CTTGAAGCTCTGAC    [48]
P._davidiana_2             CACTTCTTC-AATGCCTTTTGTTTTTCTTCTTTCA-CTTGACGCTCTGAC    [48]
P._davidiana_1             CACTTCTTC-AATGCCTTTTGTTTTTCTTCTTTCA-CTTGAAGCTCTGAC    [48]
P._davidiana_3             CACTTCTTC-AATGCCTTTTGTTTTTCTTCTTTCA-CTTGAAGCTCTGAC    [48]
P._tremuloides_1           CACTTCTTCCATTGCCTTTTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [50]
P._tremuloides_3           ----TCTTC-ATTGCCTTTTGTGTTTCTTCTTTGAACTTGAAGCTCTGAC    [45]
P._tremuloides_2           CACTTCTTC-ATTGCCTTTTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [49]
P._grandidentata_2         ---TTCTTC-ATTGCCTTTTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [46]
P._grandidentata_4         ---TTCTTC-ATTGCCTTTTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [46]
P._grandidentata_3         --CTTCTTC-ATTGCCTTTTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [47]
P._cathayana               CACTTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [49]
P._laurifolia_1            CCCTTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [49]
P._balsamifera_2           CACTTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [49]
P._balsamifera_3           --CTTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [47]
P._trichocarpa_1           CACTTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [49]
P._balsamifera_1           CACTTCTTCCATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [50]
P._simonii                 CACTTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [49]
P._laurifolia_2            CACTTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [49]
P._szechuanica             CACTTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [49]
P._trichocarpa_2           -ACTTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [48]
P._angustifolia            ---TTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [46]
P._tristis                 CACTTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [49]
P._nigra_1                 CACTTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [49]
P._nigra_3                 CACTTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [49]
P._nigra_4                 ---TTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [46]
P._nigra_5                 ---TTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTTTGAC    [46]
P._nigra_2                 --CTTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [47]
P._maximowiczii_1          ---TTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGACGCTCTGAC    [46]
P._maximowiczii_2          ---TTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [46]
P._deltoides_1             CACTTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [49]
P._deltoides_3             ---TTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [46]
P._deltoides_2             CACTTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [49]
P._sargentii               CACTTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [49]
P._angulata                ---TTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [46]
P._fremontii               CACTTCTTC-ATTGCCTTCTGTTTTTCTTCTTTGAACTTGAAGCTCTGAC    [49]
S._integra                 CACTTCTTC-ATTGCCTTTTGCTTTTCTTCTTTGAACTTGAAGCTTCCAC    [49]
S._nigra                   --CTTCTTCCATTGCCTTTTGCTTTTCTTCTTTGAACTTGAAGCTTCCAC    [48]

[                                  60         70         80         90        100]
[                                  .          .          .          .          .]

P._alba_1                  TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTCACCCCATTTTGTG    [88]
P._alba_2                  TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTCACCCCATTTTGTG    [99]
P._alba_3                  TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTCACCCCATTTTGTG    [97]
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P._tremula_1          TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTCACCCCATTTTGTG   [98]
P._tremula_2          TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTCACCCCATTTTGTG   [98]
P._davidiana_5        TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTCACCCCATTTTGTG   [98]
P._davidiana_4        TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTCACCCCATTTTGTG   [98]
P._davidiana_2        TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTCACCCCATTTTGTG   [98]
P._davidiana_1        TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTCACCCCATTTTGTG   [98]
P._davidiana_3        TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTCACCCCATTTTGTG   [98]
P._tremuloides_1      TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTCACCCCATTTTGTG   [100]
P._tremuloides_3      TGCCTTCTCCCCTCCAGCAACTCAAAGAACTTGGTTCACCCCATTTTGTG   [95]
P._tremuloides_2      TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTCACCCCATTTTGTG   [99]
P._grandidentata_2    TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTCACCCCATTTTGTG   [96]
P._grandidentata_4    TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTCACCCCATTTTGTG   [96]
P._grandidentata_3    TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTCACCCCATTTTGTG   [97]
P._cathayana          TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTT-ACCCCATTTTGTG   [98]
P._laurifolia_1       TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTG   [99]
P._balsamifera_2      TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTG   [99]
P._balsamifera_3      TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTG   [97]
P._trichocarpa_1      TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTG   [99]
P._balsamifera_1      TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTG   [100]
P._simonii            TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTG   [99]
P._laurifolia_2       TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTG   [99]
P._szechuanica        TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGCTTTACCCCATTTTGTG   [99]
P._trichocarpa_2      TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTG   [98]
P._angustifolia       TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTT-CCCCATTTTGTG   [95]
P._tristis            TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTG   [99]
P._nigra_1            TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTG   [99]
P._nigra_3            TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTG   [99]
P._nigra_4            TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTG   [96]
P._nigra_5            TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTG   [96]
P._nigra_2            TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTG   [97]
P._maximowiczii_1     TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGCTTTACCCCATTTTGTG   [96]
P._maximowiczii_2     TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTG   [96]
P._deltoides_1        TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTT   [99]
P._deltoides_3        TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTT   [96]
P._deltoides_2        TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTG   [99]
P._sargentii          TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTG   [99]
P._angulata           TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTG   [96]
P._fremontii          TGCCTTCACCCCTCCAGCAACTCAAAGAACTTGGTTTACCCCATTTTGTG   [99]
S._integra            TGCCTCCACCCCTC---------AAAGAACTTGGTTTACCCCATTCTGTG   [90]
S._nigra              TGCCTTCACCCCTC---------AAAGAACTTGGTTTACCCCATTCTGTG   [89]

[                              110       120       130       140       150]
[                              .         .         .         .         .]

P._alba_1             CCATCACCACACTCCTC------ATCATCATCAATAAAAGCCACACCACC   [132]
P._alba_2             CCATCACCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [143]
P._alba_3             CCATCACCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [141]
P._tremula_1          TCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [142]
P._tremula_2          TCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [142]
P._davidiana_5        TCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [142]
P._davidiana_4        TCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [142]
P._davidiana_2        TCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [142]
P._davidiana_1        TCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [142]
P._davidiana_3        TCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACT   [142]
P._tremuloides_1      TCATCGCCACACTCCTC------NTCGTCATCAATAAAAGCCACACCACC   [144]
P._tremuloides_3      TCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [139]
P._tremuloides_2      TCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [143]
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P._grandidentata_2    TCATCGCCACACTCCTC------ATCATCATCAATAAAAGCCACACCACC   [140]
P._grandidentata_4    TCATCGCCACACTCCTC------ATCATCATCAATAAAAGCCACACCACC   [140]
P._grandidentata_3    TCATCGCCACACTCCTC------ATCATCATCAATAAAAGCCACACCACC   [141]
P._cathayana          CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [142]
P._laurifolia_1       CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [143]
P._balsamifera_2      CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [143]
P._balsamifera_3      CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [141]
P._trichocarpa_1      CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [143]
P._balsamifera_1      CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [144]
P._simonii            CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [143]
P._laurifolia_2       CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [143]
P._szechuanica        CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [143]
P._trichocarpa_2      CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [142]
P._angustifolia       CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [139]
P._tristis            CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [143]
P._nigra_1            CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [143]
P._nigra_3            CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [143]
P._nigra_4            CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [140]
P._nigra_5            CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [140]
P._nigra_2            CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [141]
P._maximowiczii_1     CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [140]
P._maximowiczii_2     CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [140]
P._deltoides_1        CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [143]
P._deltoides_3        CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [140]
P._deltoides_2        CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [143]
P._sargentii          CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [143]
P._angulata           CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [140]
P._fremontii          CCATCGCCACACTCCTC------ATCGTCATCAATAAAAGCCACACCACC   [143]
S._integra            CCATCACCACACTCCTCCTCATCATCATCATCAATAAAAGCCACACCACC   [140]
S._nigra              CCATCACCACACTCATC------ATCATCATCAATAAAAACAACAACACC   [133]

[                              160       170       180       190       200]
[                              .         .         .         .         .]

P._alba_1             GTCCAAATCATCCTCTGGAACTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [182]
P._alba_2             GTCCAAATCATCCTCTGGAACTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [193]
P._alba_3             GTCCAAATCATCCTCTGGAACTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [191]
P._tremula_1          GTCCAAATCATCCTCTGGAACTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [192]
P._tremula_2          GTCCAAATCATCCTCTGGAACTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [192]
P._davidiana_5        GTCCAAATCATCCTCTGGAACTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [192]
P._davidiana_4        GTCCAAATCATCCTCTGGAACTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [192]
P._davidiana_2        GTCCAAATCATCCTCTGGAACTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [192]
P._davidiana_1        GTCCAAATCATCCTCTGGAACTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [192]
P._davidiana_3        GTCCAAATCATCCTCTGGAACTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [192]
P._tremuloides_1      GTCNAAATCATCCTCTGGAACTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [194]
P._tremuloides_3      GTCTAAATCATCCTCTGGAACTTCTGCTTCGGGAGGCTCCCAAACCCGTG   [189]
P._tremuloides_2      GTCTAAATCATCCTCTGGAACTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [193]
P._grandidentata_2    GTCCAAATCATCCTCTGGAACTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [190]
P._grandidentata_4    GTCCAAATCATCCTCTGGAACTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [190]
P._grandidentata_3    GTCCAAATCATCCTCTGGAACTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [191]
P._cathayana          GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [192]
P._laurifolia_1       GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [193]
P._balsamifera_2      GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [193]
P._balsamifera_3      GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [191]
P._trichocarpa_1      GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [193]
P._balsamifera_1      GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [194]
P._simonii            GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [193]
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P._laurifolia_2        GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [193]
P._szechuanica         GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [193]
P._trichocarpa_2       GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [192]
P._angustifolia        GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCATG   [189]
P._tristis             GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [193]
P._nigra_1             GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [193]
P._nigra_3             GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [193]
P._nigra_4             GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [190]
P._nigra_5             GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [190]
P._nigra_2             GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [191]
P._maximowiczii_1      GTCCAAATCATTCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [190]
P._maximowiczii_2      GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [190]
P._deltoides_1         GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [193]
P._deltoides_3         GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [190]
P._deltoides_2         GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [193]
P._sargentii           GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [193]
P._angulata            GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [190]
P._fremontii           GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [193]
S._integra             GTCCAAATCATCCTCTGGATCTTCTGCTTCAGGAGGCTCCCAAACCCGTG   [190]
S._nigra               ATCGTAATAATCATCTGGAGGATCTGCTGCTTGAGGATGCTCAAAAACCG   [183]

[                              210        220        230        240        250]
[                              .          .          .          .          .]

P._alba_1                CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [232]
P._alba_2                CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [243]
P._alba_3                CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [241]
P._tremula_1             CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [242]
P._tremula_2             CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [242]
P._davidiana_5           CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [242]
P._davidiana_4           CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [242]
P._davidiana_2           CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [242]
P._davidiana_1           CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [242]
P._davidiana_3           CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [242]
P._tremuloides_1         CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [244]
P._tremuloides_3         CATCTACTTCGTCGTCGCTGATTTGAGAAATATCATTTCCTTCATTTGAA   [239]
P._tremuloides_2         CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [243]
P._grandidentata_2       CATCTACTTCATCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [240]
P._grandidentata_4       CATCTACTTCATCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [240]
P._grandidentata_3       CATCTACTTCATCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [241]
P._cathayana             CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [242]
P._laurifolia_1          CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [243]
P._balsamifera_2         CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [243]
P._balsamifera_3         CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [241]
P._trichocarpa_1         CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [243]
P._balsamifera_1         CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [244]
P._simonii               CATCTACTTCGTCATCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [243]
P._laurifolia_2          CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [243]
P._szechuanica           CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [243]
P._trichocarpa_2         CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [242]
P._angustifolia          CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [239]
P._tristis               CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [243]
P._nigra_1               CATCTGCTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [243]
P._nigra_3               CATCTGCTTCGTCGTCACTGATTTGAGAAATATAATTTCCTTCATTTGAA   [243]
P._nigra_4               CATCTGCTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [240]
P._nigra_5               CATCTGCTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [240]
P._nigra_2               CATCTGCTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [241]
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P._maximowiczii_1     CATCTACTTCGTCATCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [240]
P._maximowiczii_2     CATCTACTTCGTCATCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [240]
P._deltoides_1        CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTGCTTCATCTGAA   [243]
P._deltoides_3        CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTGCTTCATCTGAA   [240]
P._deltoides_2        CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTGCTTCATCTGAA   [243]
P._sargentii          CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTGCTTCATCTGAA   [243]
P._angulata           CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [240]
P._fremontii          CATCTACTTCGTCGTCACTGATTTGAGAAATATCATTTCCTTCATTTGAA   [243]
S._integra            CATCTACTTCGTTGTCACTAATTTGAGAAATATCACTTCCTTCATTTGAA   [240]
S._nigra              CAGCATCTTCGTTGTTGTTGATAAGATAAAAAATATTTCTTTCATTTGAA   [233]

[                              260        270        280        290        300]
[                              .          .          .          .          .]

P._alba_1             GATCCAACACTGTTCACAGAAGAGTTACCTTTTGATTCTTGATCGTGAAC   [282]
P._alba_2             GATCCAACACTGTTCACAGAAGAGTTACCTTTTGATTCTTGATCGTGAAC   [293]
P._alba_3             GATCCAACACTGTTCACAGAAGAGTTACCTTTTGATTCTTGATCGTGAAC   [291]
P._tremula_1          GATCCAACACTGTTCACAAAAGAGTTACCTTTTGATTCTTGATTGTGAAC   [292]
P._tremula_2          GATCCAACACTGTTCACAAAAGAGTTACCTTTTGATTCTTGATTGTGAAC   [292]
P._davidiana_5        GATCCAACACTGTTCACAGAAGAGTTACCTTTTGATTCTTGATTGTGAAC   [292]
P._davidiana_4        GATCCAACACTGTTCACAGAAGAGTTACCTTTTGATTCTTGATTGTGAAC   [292]
P._davidiana_2        GATCCAACACTGTTCACAGAAGAGTTACCTTTTGATTCTTGATTGTGAAC   [292]
P._davidiana_1        GATCCAACACTGTTCACAGAAGAGTTACCTTTTGATTCTTGATCGTGAAC   [292]
P._davidiana_3        TATCCAACACTGTTCACAGAAGAGTTACCTTTTGATTCTTGATCGTGAAC   [292]
P._tremuloides_1      GATCCAACACTGTTCACAGAAGAGTTACCTTTTGATTCTTGATCATGAAC   [294]
P._tremuloides_3      GATCCAACACTGTTCACATAAGAGTTACCTTTTGATTCTTGATCATGAAC   [289]
P._tremuloides_2      GATCCAACACTGTTCACAGAAGAGTTACCTTTTGATTCTTGATCATGAAC   [293]
P._grandidentata_2    GATCCAACACTGTTCACAGAAGAGTTACCTTTTGATTCTTGATCGTGAAC   [290]
P._grandidentata_4    GATCCAACACTGTTCACAGAAGAGTTACCTTTTGATTCTTGATCGTGAAC   [290]
P._grandidentata_3    GATCCAACACTGTTCACAGAAGAGTTACCTTTTGATTCTTGATCGTGAAC   [291]
P._cathayana          AATCCAACACTGTTCACAGAAGAGTTACCTTCTGATTCTTGATCATTAAC   [292]
P._laurifolia_1       GATCCAACACTGTTCACAGAAGAGTTACCTTCTGATTCTTGATCATTAAC   [293]
P._balsamifera_2      AATCCAACACTGTTCGCAGAAGAGTTACCTTCTGATTCTTGATCGTTAAC   [293]
P._balsamifera_3      AATCCAACACTGTTCGCAGAAGAGTTACCTTCTGATTCTTGATCGTTAAC   [291]
P._trichocarpa_1      AATCCAACACTGTTCGCAGAAGAGTTACCTTCTGATTCTTGATCGTTAAC   [293]
P._balsamifera_1      NATCCAACACTGTTCNCAGAAGAGTTACCTTCTGATTCTTGATCGTTAAC   [294]
P._simonii            GATCCAACACTGTTCACAGAAGAGTTACCTTCTGATTCTTGATCATTAAC   [293]
P._laurifolia_2       GATCCAACACTGTTCACAGAAGAGTTACCTTCTGATTCTTGATCATTAAC   [293]
P._szechuanica        AATCCAACACTGTTCACAGAAGAGTTACCTTCTGATTCTTGATCGTTAAC   [293]
P._trichocarpa_2      AATCCAACACTGTTCACAGAAGAGTTACCTTCTGATTCTTGATCGTTAAC   [292]
P._angustifolia       AATCCAACACTGTTCACTGAAGAGTTACCTTCTGATTCTTGATCGTTAAC   [289]
P._tristis            GATCCAACACTGTTCACAGAAGAGTTACCTTCTGATTCTTGATCGTTAAC   [293]
P._nigra_1            GATCCAACACTGTTCACAGAAGAGTTACCTTCTGATTCTCGATCGTTAAC   [293]
P._nigra_3            GATCCAACACTGTTCACAGAAGAGTTACCTTCTGATTCTCGATCGTTAAC   [293]
P._nigra_4            GATCCAACACTGTTCACAGAAGAGTTACCTTCTGATTCTCGATCGTTAAC   [290]
P._nigra_5            GATCCAACACTGTTCACAGAAGAGTTACCTTCTGATTCTCGATCGTTAAC   [290]
P._nigra_2            GATCCAACACTGTTCACAGAAGAGTTACCTTCTGATTCTCGATCGTTAAC   [291]
P._maximowiczii_1     GATCCAACACTGTTCACAGAAGAGTTACCTTCTGATTCTTGATCGTGAAC   [290]
P._maximowiczii_2     GATCCAACACTGTTCACAGAAGAGTTACCTTCTGATTCTTGATCGTTAAC   [290]
P._deltoides_1        GATCCAACACTGTTCACAGAAGAGTTACCTTCTGATTCTTGATCGTGAAC   [293]
P._deltoides_3        GATCCAACACTGTTCACAGAAGAGTTACCTTCTGATTCTTGATCGTGAAC   [290]
P._deltoides_2        GATCCAACACTGTTCACAGAAGAGTTACCTTCTGATTCTTGATCGTGAAC   [293]
P._sargentii          GATCCAACACTGTTCACAGAAGAGTTACCTTCTGATTCTTGATCGTGAAC   [293]
P._angulata           GATCCAACACTGTTCACAGAAGAGTTACCTTCTGATTCTTGATCGTGAAC   [290]
P._fremontii          GATCCAACACTGTTCACAGAAGAGTTACCTTCTGATTCTTGATCGTGAAC   [293]
S._integra            GATCCAACACTGTTCTCAGAAGAGTTTCCTTTTGATTCTTGATCATGAAT   [290]
S._nigra              GAACATACAACGTTGTTATAAGAATATTTTTTTGATTATTGATGATGAAG   [283]
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[                          310        320        330        340        350]
[                          .          .          .          .          .]

P._alba_1                  AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [332]
P._alba_2                  AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [343]
P._alba_3                  AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [341]
P._tremula_1               AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [342]
P._tremula_2               AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [342]
P._davidiana_5             AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [342]
P._davidiana_4             AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [342]
P._davidiana_2             AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [342]
P._davidiana_1             AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [342]
P._davidiana_3             AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [342]
P._tremuloides_1           AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [344]
P._tremuloides_3           AAGTTCCACAATTTCCACATCTCTCACTGTGTCTCTATCACTGCCATTTT    [339]
P._tremuloides_2           AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [343]
P._grandidentata_2         AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [340]
P._grandidentata_4         AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [340]
P._grandidentata_3         AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [341]
P._cathayana               AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [342]
P._laurifolia_1            AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [343]
P._balsamifera_2           AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [343]
P._balsamifera_3           AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [341]
P._trichocarpa_1           AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [343]
P._balsamifera_1           AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [344]
P._simonii                 AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [343]
P._laurifolia_2            AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [343]
P._szechuanica             AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [343]
P._trichocarpa_2           AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [342]
P._angustifolia            AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [339]
P._tristis                 AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [343]
P._nigra_1                 AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [343]
P._nigra_3                 AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [343]
P._nigra_4                 AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [340]
P._nigra_5                 AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [340]
P._nigra_2                 AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [341]
P._maximowiczii_1          AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [340]
P._maximowiczii_2          AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [340]
P._deltoides_1             AAGTTCCACAATTTCTACATCTCTCACTTTGTCTCTATCACTGCCATTAT    [343]
P._deltoides_3             AAGTTCCACAATTTCTACATCTCTCACTTTGTCTCTATCACTGCCATTAT    [340]
P._deltoides_2             AAGTTCCACAATTTCTACATCTCTCACTTTGTCTCTATCACTGCCATTAT    [343]
P._sargentii               AAGTTCCACAATTTCTACATCTCTCACTTTGTCTCTATCACTGCCATTAT    [343]
P._angulata                AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [340]
P._fremontii               AAGTTCCACAATTTCTACATCTCTCACTGTGTCTCTATCACTGCCATTAT    [343]
S._integra                 AAGTTCCACAATTTCTATATCTCTCACTGTGTCTCTATCACTGCCATTAT    [340]
S._nigra                   A--------------------------------------------------    [284]

[                          360        370        380        390        400]
[                          .          .          .          .          .]

P._alba_1                  GGATATCCTTCTCCGTCACTTTATGCAATCCATCCACTCTATTTACAGGA    [382]
P._alba_2                  GGATATCCTTCTCCGTCACTTTATGCAATCCATCCACTCTATTTACAGGA    [393]
P._alba_3                  GGATATCCTTCTCCGTCACTTTATGCAATCCATCCACTCTATTTACAGGA    [391]
P._tremula_1               GGATATCCTTCTCCATCACTTTATGCAATCCATCCACTCTATTTACAGGA    [392]
P._tremula_2               GGATATCCTTCTCCGTCACTTTATGCAATCCATCCACTCTATTTACAGGA    [392]
P._davidiana_5             GGATATCCTTCTCCGTCACTTTATGCAATCCATCCACTCTATTTACAGGA    [392]
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P._davidiana_4        GGATATCCTTCTCCGTCACTTTATGCAATCCATCCACTCTATTTACAGGA   [392]
P._davidiana_2        GGATATCCTTCTCCGTCACTTTATGCAATCCATCCACTCTATTTACAGGA   [392]
P._davidiana_1        GGATATCCTTCTCCATCACTTTATGCAAACCATCCACTCTATTTACAGGA   [392]
P._davidiana_3        GGACATCCTTCTCCGTCACTTTATGCAATCCATCCACTCTATTTACAGGA   [392]
P._tremuloides_1      GGATATCCTTCTCCGTCACTTTATGCAATCCATCCACTCTATTTACAGGA   [394]
P._tremuloides_3      GGATATCCTTCTCCGTCACTTTATGCAATCCATCCACTCTATTTACAGGA   [389]
P._tremuloides_2      GGATATCCTTCTCCGTCACTTTATGCAATCCATCCACTCTATTTACAGGA   [393]
P._grandidentata_2    GGATATCCTTCTCTGTCACTTTATGCAATCCATCCACTCTATTTACAGGA   [390]
P._grandidentata_4    GGATATCCTTCTCTGTCACTTTATGCAATCCATCCACTCTATTTACAGGA   [390]
P._grandidentata_3    GGATATCCTTCTCTGTCACTTTATGCAATCCATCCACTCTATTTACAGGA   [391]
P._cathayana          GGCTATCCTTCCCTGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [392]
P._laurifolia_1       GGCTATCCTTCCCTGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [393]
P._balsamifera_2      GGCTATCCTTCCCTGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [393]
P._balsamifera_3      GGCTATCCTTCCCTGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [391]
P._trichocarpa_1      GGCTATCCTTCCCTGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [393]
P._balsamifera_1      GGCTATCCTTCCCTGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [394]
P._simonii            GGCTATCCTTCCCTGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [393]
P._laurifolia_2       GGCTATCCTTCCCTGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [393]
P._szechuanica        GGCTATCCTTCCCTGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [393]
P._trichocarpa_2      GGCTATCCTTCCCTGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [392]
P._angustifolia       GGCTATCCTTCCCTGTCACTTTATGTAATCCATCCACTCTATCTACAGGA   [389]
P._tristis            GGCTATCCTTCCCTGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [393]
P._nigra_1            GGCTATCCTTCCCCGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [393]
P._nigra_3            GGCTATCCTTCCCCGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [393]
P._nigra_4            GGCTATCCTTCCCCGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [390]
P._nigra_5            GGCTATCCTTCCCCGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [390]
P._nigra_2            GGCTATCCTTCCCCGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [391]
P._maximowiczii_1     GGCTATCCTTCCCCGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [390]
P._maximowiczii_2     GGCTATCCTTCCCCGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [390]
P._deltoides_1        GGCTATCCTTCTCCGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [393]
P._deltoides_3        GGCTATCCTTCTCCGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [390]
P._deltoides_2        GGCTATCCTTCTCCGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [393]
P._sargentii          GGCTATCCTTCTCCGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [393]
P._angulata           GGCTATCCTTCTCCGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [390]
P._fremontii          GGCTATCCTTCCCCGTCACTTTATGTAATCCATCCACTCTATTTACAGGA   [393]
S._integra            GGCTATCCTTTTCCATCACTTTATGTAATCCATCAACTCTATTTACAGGA   [390]
S._nigra              --------------------------------------------------   [284]

[                              410]
[                              .]

P._alba_1             GCTTCC----   [388]
P._alba_2             GCTTCCAAT-   [402]
P._alba_3             GCTTCCAA--   [399]
P._tremula_1          GCTTCCAATC   [402]
P._tremula_2          GCTTCCA---   [399]
P._davidiana_5        GCTTCCAATC   [402]
P._davidiana_4        GCTTCCAATC   [402]
P._davidiana_2        GCTTCCAATC   [402]
P._davidiana_1        GCTTCCAATC   [402]
P._davidiana_3        GCTTCCAATC   [402]
P._tremuloides_1      GCTTCCAATC   [404]
P._tremuloides_3      GCTTCCAATC   [399]
P._tremuloides_2      GCTTCCAA--   [401]
P._grandidentata_2    GCTTCCA---   [397]
P._grandidentata_4    GCTTCC----   [396]
P._grandidentata_3    GCTT------   [395]
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P._cathayana            GCTTCCAATC   [402]
P._laurifolia_1         GCTTCCAATC   [403]
P._balsamifera_2        GCTTCCAATT   [403]
P._balsamifera_3        GCT-------   [394]
P._trichocarpa_1        GCTTCCAATC   [403]
P._balsamifera_1        GCTTCCAA--   [402]
P._simonii              GCTT------   [397]
P._laurifolia_2         GCTTCCAA--   [401]
P._szechuanica          GCTTCCAA--   [401]
P._trichocarpa_2        GCTTCCAAT-   [401]
P._angustifolia         GCTTCCAAT-   [398]
P._tristis              GCTTCCAATC   [403]
P._nigra_1              GCTTCCAATC   [403]
P._nigra_3              GCTTCCAA--   [401]
P._nigra_4              GCTTCCAATC   [400]
P._nigra_5              GCTTCCAAT-   [399]
P._nigra_2              GCTTCCA---   [398]
P._maximowiczii_1       GCTTCCAATC   [400]
P._maximowiczii_2       GCTTCCAAT-   [399]
P._deltoides_1          GCTTCCAATC   [403]
P._deltoides_3          GCTTC-----   [395]
P._deltoides_2          GCTTCCAA--   [401]
P._sargentii            GCTTCCAATC   [403]
P._angulata             GCTTCCAATC   [400]
P._fremontii            GCTTCCAATC   [403]
S._integra              GCTTCCAATC   [400]
S._nigra                ----------   [284]
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Appendix 2. 3 Nucleotide sequence data matrix of SCISSR fragment PIS5A1.

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[                          10         20         30         40         50]
[                          .          .          .          .          .]

P._alba_1                  AAAAATAGTCGAGGGACTAATCGAACTTCT-TCTTAAGCAAATCAAGAAA   [49]
P._alba_3                  AAAAATAGTCGAGGGACTAATCGAACTTCT-TCTTAAGCAAATCAAGAAA   [49]
P._davidiana_4             --AAATAGTCGAGGGACTAATCAAACTTCTGTCTTAAACAAATCAAGAAA   [48]
P._trichocarpa_2           --------------------------------------------------   [0]
P._szechuanica             AAAAATAGTCGAGGGACTAATCGAACTTCG-TCTTAAACAAATCAAGAAA   [49]
p._maximowiczii_2          AAAAATAGTTGAGGGACTAATCGAACTTCG-TCTTAAACAAATCAAGAAA   [49]
P._simonii                 AAAAATAGTTGAGGGACTAATCGAACTTCG-TCTTAA-CAAATCAAGAAA   [48]
P._cathayana               AAAAATAGTCGAGGGACTAATCGAACTTCG-TCTTAAACAAATCAAGAAA   [49]
P._maximowiczii_1          AAAAATAGTCGAGGGACTAATCGAACTTCG-TCTTAAACAAATCAAGAAA   [49]
P._laurifolia_2            -AAAATAGTCGAGGGACTAATCGAACTTCG-TCTTAAACAAATCAAGAAA   [48]
P._nigra_1                 -AAAATAGTCGAGGGACTAATCGAACTTCG-TCTTAAACAAATCAAGAAA   [48]
P._nigra_4                 AAAAATAGTCGAGGGACTAATCGAACTTCG-TCTTATACAAATCAAGAAA   [49]
P._nigra_2                 -AAAATAGTCGAGGGACTAATCGAACTTCG-CCTTAAACAAATCAAGAAA   [48]
P._fremontii               AAAAATAGTCGAGGGACTAATCGAACTTCG-TCTTAAACAAATCAAGAAA   [49]
P._balsamifera_2           -AAAATAGTCGAGGGACTAATCGAACTTCG-TCTTAAACAAATCAAGAAA   [48]
P._balsamifera_3           -AAAATAGTCGAGGGACTAATCGAACTTCG-TCTTAAACAAATCAAGAAA   [48]
P._balsamifera_1           -AAAATAGTCGAGGGACTAATCGAACTTCG-TCTTAAACAAATCAAGAAA   [48]
P._tremuloides_2           ----------------------------------------ATCAAGAAA   [9]
P._nigra_3                 -----------------------------------------AAATCAAGAAA   [11]
P._laurifolia_1            -----TAGTCGAGGGACTAATCGAACTTCG-TCTTAAACAAATCAAGAAA   [44]
P._davidiana_1             -AAAATAGTCTAGGGACTAATCGAACTTCG-TCTTAAACAAATCAAGAAA   [48]
P._grandidentata_2         AAAAATAGTCGAGGGACTAATCGAACTTCG-TCTTAAACAAATCAAGAAA   [49]
P._grandidentata_3         -AAAATAGTCGAGGGACTAATCGAACTTCGCTCTTAAACAAATCAAGAAA   [49]
P._grandidentata_4         -AAAATAGTCGAGGGACTAATCGAACTTCG-TCTTAAACAAATCAAGAAA   [48]
P._deltoides_1             -AAAATAGTCGAGGGACTAATCGAACTTCG-TCTTAAACAAATCTAGAAA   [48]
P._sargentii               -AAAATAGTCGAGGGACTAATCGAACTTCG-TCCTATA-AAATCTAGAAA   [47]
P._deltoides_2             -AAAATAGTCGAGGGACTAATCGAACTTCG-TCTTAAACAAATCTAGAAA   [48]
P._deltoides_3             -AAAATAGTCGAGGGACTAATCGAACTTCGCTCTTAAACAAATCTAGAAA   [49]
P._angulata                -AAAATAGTCGAGGGACTAATCGAACTTCG-TCTTAAACAAATCAAGAAA   [48]
P._tremula_1               AAAAATAGTCGAGGAACTAATCGAACTTC-TTCTTAAACAAATCAAGAAA   [49]
P._nigra_5                 -AAAATAGTCGAGGGACTAATCGAACTTCG-TCTTAAACAAATCAAGAAA   [48]

[                          60         70         80         90         100]
[                          .          .          .          .          .]

P._alba_1                  GCAAGTAAACAGACCTCACTCGCCTTCACAACCTCTGC-----TGCCAGA   [94]
P._alba_3                  GCAAGTAAACAGACCTCACTCACCTTCACAACCTCTGC-----TGCCAGA   [94]
P._davidiana_4             GCAAGTAAACAGACCTCACTCACCTTCACTACCTCTGC-----TGCCAGA   [93]
P._trichocarpa_2           ----------------------------------CTGG--------CAGA   [8]
P._szechuanica             GCAAGTAAACAGACCTCACTCTCCTTCACTACCTCTGCGGCTCTGCCAGA   [99]
p._maximowiczii_2          ACAAGTAAACAGACCTCACTCTCCTTCACTACCTCTGC--------CAGA   [91]
P._simonii                 GCAAGTAAACAGACCTCACTCTCCTTCACTACCTCTGC--------CAGA   [90]
P._cathayana               GCAAGTAAACAGACCTCACTCTCCTTCACTACCTCTGC--------CAGA   [91]
P._maximowiczii_1          GCAAGTAAATAGACCTCACTCTCCTTCACTACCTCTGC--------CAGA   [91]
P._laurifolia_2            GCAAGTAAACAGACCTCACTCTCCTTCACTACCTCTGC--------CAGA   [90]
P._nigra_1                 GCAAGTAAACAGACCTCACTCTCCTTCACTACCTCTGC--------CAGA   [90]
P._nigra_4                 GCAAGTAAACAGACCTCACTCTCCTTCACTACCTCTGC--------CAGA   [91]
P._nigra_2                 GCAAGTAAACAGACCTCACTCTCCTTCACTACCTCTGC--------CAGA   [90]
P._fremontii               GCAAGTAAACAGACCTCACTCTCCTTCACTACCTCTGC--------CAGA   [91]
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P._balsamifera_2      GCAAGTAAACAGACCTCACTCTCCTTCACTACCTCTGC--------CAGA    [90]
P._balsamifera_3      GCAAGTAAACAGACCTCACTCTCCTTCACTACCTCTGC--------CAGA    [90]
P._balsamifera_1      GCAAGTAAACAGACCTCACTCTCCTTCACTACCTCTGC--------CAGA    [90]
P._tremuloides_2      GCAAGTAAACAGACCTCACTCACCTTCACTACCTCTGC--------CAGA    [51]
P._nigra_3            GCAAGTAAACAGACCTCACTCTCCTTCACTACCTCTGC--------CAGA    [53]
P._laurifolia_1       GCAAGTAAACAGACCTCACTCTCCTTCACTACCTCTGC--------CAGA    [86]
P._davidiana_1        GCAAGTAAACAGACCTCACTCACCTTCACTACCTCTGC--------CAGA    [90]
P._grandidentata_2    GCAAGTAAACAGACCTCACTCACCTTCACTACCTCTGC--------CAGA    [91]
P._grandidentata_3    GCAAGTAAACAGACCTCACTCACCTTCACTACCTCTGC--------CAGA    [91]
P._grandidentata_4    GCAAGTAAACAGACCTCACTCACCTTCACTACCTCTGC--------CAGA    [90]
P._deltoides_1        GC----AAACAGACCTTGCTCTGCTTCACTACCTCTGC--------CAGA    [86]
P._sargentii          GC----AAACAGACCTTGCTCTGCTTCACCTCCTCTGC--------CAGA    [85]
P._deltoides_2        GC----AAACAGACCTTGCTCTGCTTCACTACCTCTGC--------CAGA    [86]
P._deltoides_3        GC----AAACAGACCTTGCTCTGCTTCACTACCTCTGC--------CAGA    [87]
P._angulata           GCAAGTAAACAGACCTCACTCTCCTTCACTACCTCTGC--------CAGA    [90]
P._tremula_1          GCAAGTAAACAGACCTCACTCACCTTCACTACCTCTGC-----TGCCAGA    [94]
P._nigra_5            GCAAGTAAACAGACCTCACTCTCCTTCACTACCTCTG--------CCAGA    [90]

[                              110       120       130       140       150]
[                              .         .         .         .         .]

P._alba_1             TATTCTAATTTTTTCTCTACGGATCGATCA-GTTGCTGAAACCCTAGATC    [143]
P._alba_3             TATTCTAATTTTTTCTCTACGGATCGATCA-GTTGCTGAAACCCTAGATC    [143]
P._davidiana_4        TATTCTAATTTTTTCTCTACGGATCGACCA-GTTGCTGAAACCCTAGATC    [142]
P._trichocarpa_2      TATTCTAATTTTT-CTCTACGGATCGATCAAGTTGCTGAAACCCTAGATC    [57]
P._szechuanica        TATTCTAATTTTT-CTCTACGGATCGATCA-GTTGCTGAAACCCTAGATC    [147]
p._maximowiczii_2     TATTCTAATTTTT-CTCTACGGATCGATCA-GTTGCTGAAACCCTAGATC    [139]
P._simonii            TATTCTAATTTTT-CTCTACGGATCGATCA-GTTGCTGAAACCCTAGATC    [138]
P._cathayana          TATTCTAATTTTT-CTTTACGGATCGATCA-GTTGCTGAAACCCTAGATC    [139]
P._maximowiczii_1     TATTCTAATTTTTTCTCTACGGATCGATCA-GTTGCTGAAACCCTAGATC    [140]
P._laurifolia_2       TATTCTAATTTTTTCTCTACGGATCGATCA-GTTGCTGAAACCCTAGATC    [139]
P._nigra_1            TATTCTAATTTTT-CTCTACGGATCGATCA-GTTGCTGAAACCCTAGATC    [138]
P._nigra_4            TATTCTAATTTTT-CTCTACGGATCGATCA-GTTGCTGAAACCCTAGATC    [139]
P._nigra_2            TATTCTAATTTTT-CTCTACGGATCGATCA-GTTGCTGAAACCCTAGATC    [138]
P._fremontii          TATTCTAATTTTT-CTCTACGGATCGATCA-GTTGCTGAAACCCTAGATC    [139]
P._balsamifera_2      TATTCTAATTTTT-CTCTACGGATCGATCA-GTTGCTGAAACCCTAGATC    [138]
P._balsamifera_3      TATTCTAATTTTT-CTCTACGGATCGATCA-GTTGCTGAAACCCTAGATC    [138]
P._balsamifera_1      TATTCTAATTTTT-CTCTACGGATCGATCA-GTTGCTGAAACCCTAGATC    [138]
P._tremuloides_2      TATTCTATTTTTT-CTCTACGGATTGATCA-GTTGCTGAAACCCTAGATC    [99]
P._nigra_3            TATTCTAATTTTT-CTCTACGGATCGATCA-GTTGCTGAAACCCTAGATC    [101]
P._laurifolia_1       TATTCTAATTTTTTCTCTACGGATCGATCA-GTTGCTGAAACCCTAGATC    [135]
P._davidiana_1        TATTCTAATTTTT-CTCTACGGATTGATCA-GTTGCTGAAACCCTAGATC    [138]
P._grandidentata_2    TATTCTAATTTTT-CTCTACGGATTGATCA-GTTGCTGAAACCCTGGATC    [139]
P._grandidentata_3    TATTCTAATTTTT-CTCTACGGATTGATCA-GTTGCTGAAACCCTAGATC    [139]
P._grandidentata_4    TATTCTAATTTTT-CTCTACGGATTGATCA-GTTGCTGAAACCCTAGATC    [138]
P._deltoides_1        TATTCTATTTTTT-CTCTACGGTTCGATCA-GTTGCTGAAACCCTAGATC    [134]
P._sargentii          TATTCTATTTTTT-CTCTACGGTTCGATCA-GTTGCTGAAACCCTAGATC    [133]
P._deltoides_2        TATTCTATTTTTT-CTCTACGGTTCGATCA-GTTGCTGAAACCCTAGATC    [134]
P._deltoides_3        TATTCTATTTTTT-CTCTACGGTTCGATCA-GTTGCTGAAACCCTAGATC    [135]
P._angulata           TATTCTAATTTTT-CTCTACGGATCGATCA-GTTGCTGAAACCCTAGATC    [138]
P._tremula_1          TATTCTATTTTTTTCTCTACGGATCGATCA-GTTGCTGAAACCCTAGATC    [143]
P._nigra_5            TATTCTAATTTTT-CTCTACGGATCGATCA-GTTGCTGAAACCCTAGATC    [138]

[                              160       170       180       190       200]
[                              .         .         .         .         .]

P._alba_1             C-----GATCCGATCCAAAAAACGATGTCTTCAAGGAAACAGCAAGAGCG    [188]
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P._alba_3              C-----GATCCGATCCGAAAAACGATGTCGTCAAGGAAACAGCAAGAGCG   [188]
P._davidiana_4         C-----GATCCTATCCAAAAAACGATGTCGTCAAGGAAACAGCGAGAGCG   [187]
P._trichocarpa_2       CAATCCGATCCGATCCAAAAAACGATGTCGTCAAGGAAACAGCGAGAGCG   [107]
P._szechuanica         C-----GATCCGATCCAAAAAACGATGTCGTCAAGGAAACAGCGAGAGCG   [192]
p._maximowiczii_2      C-----GATCCGATCCAAAAAACGATGTCGTCAGGGAAACAGCGAGAGCG   [184]
P._simonii             C-----GATCCGATCCAAAAAACGATGTCATCAGGGAAACAGCGAGAGCG   [183]
P._cathayana           C-----GATCCAATCCAAAAAACGATGTCGTCAGGGAAACAGCGAGAGCG   [184]
P._maximowiczii_1      C-----GATCCGATCCCAAAAACGATGTCGTCAGGGAAACAGCGAGAGCG   [185]
P._laurifolia_2        C-----GATCCGATCCCAAAAACGATGTCGTCAGGGAAACAGCGAGAGCG   [184]
P._nigra_1             C-----GATCCGATCCAAAAAACGATGTCGTCAAGGAAACAGCGAGAGCG   [183]
P._nigra_4             C-----GATCCGATCCAAAAAACGATGTCGTCAAGGAAACAGCGAGAGCG   [184]
P._nigra_2             C-----GATCC-----AAAAAACGATGTCATCAGGGAAACGGCGAGAGCG   [178]
P._fremontii           C-----GATCCGATCCAAAAAACGATGTCGTCAAGGAAACAGCGAGAGCG   [184]
P._balsamifera_2       C-----AATCCGATCCAAAAAACGATGTCGTCAAGGAAACAGCGAGAGCG   [183]
P._balsamifera_3       C-----AATCCGATCCAAAAAACGATGTCGTCAAGGAAACAGCGAGAGCG   [183]
P._balsamifera_1       C-----AATCCGATCCAAAAAACGATGTCGTCAAGGAAACAGCGAGAGCG   [183]
P._tremuloides_2       C-----GATCCGATCCAAAAAACGATGTCGTCAGGGAAACAGCGAGAGCG   [144]
P._nigra_3             C-----GATCCGATCCAAAAAACGATGTCGTCAAGGAAACAGCGAGAGCG   [146]
P._laurifolia_1        C-----GATCCGATCCCAAAAACGATGTCGTCAGGGAAACAGCGAGAGCG   [180]
P._davidiana_1         C-----GATCCGATCCAAAAAACGATGTCGTCAGGGAAACAGCGAGAGCG   [183]
P._grandidentata_2     C-----GATCCGATCCAAAAAACGATGTCGTCAGGGAAACAGCGAGAGCG   [184]
P._grandidentata_3     C-----GATCCGATCCAAAAAACGATGTCGTCAGGGAAACAGCGAGAGCG   [184]
P._grandidentata_4     C-----GATCCGATCCAAAAAACGATGTCGTCAGGGAAACAGCGAGAGCG   [183]
P._deltoides_1         C-----GATCCGATCCAAAAAACGATGTCGTCAAGGAAACAGCGAGAGCG   [179]
P._sargentii           C-----GATCCGATCCAAAAAACGATGTCGTCAAGGAAACAGCGAGAGCG   [178]
P._deltoides_2         C-----GATCCGATCCAAAAAACGATGTCGTCAAGGAAACAGCGAGAGCG   [179]
P._deltoides_3         C-----GATCCGATCCAAAAAACGATGTCGTCAAGGAAACAGCGAGAGCG   [180]
P._angulata            C-----GATCCGATCCAAAAAACGATGTCGTCAAGGAAACAGCGAGAGCG   [183]
P._tremula_1           C-----GATCCGATCCAAAAAACGATGTCGTCAAGGAAACAGCGAGAGCG   [188]
P._nigra_5             C-----GATCCGATCCAAAAAACGATGTCGTCAAGGAAACAGCGAGAGCG   [183]

[                              210        220        230        240        250]
[                              .          .          .          .          .]

P._alba_1              TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [238]
P._alba_3              TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [238]
P._davidiana_4         TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [237]
P._trichocarpa_2       TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [157]
P._szechuanica         TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [242]
p._maximowiczii_2      TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [234]
P._simonii             TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [233]
P._cathayana           TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [234]
P._maximowiczii_1      TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [235]
P._laurifolia_2        TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [234]
P._nigra_1             TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [233]
P._nigra_4             TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [234]
P._nigra_2             TCGAATGCTATAATCAATCTACTCAATGATCGGTCTCGTCTTCATCCTTG   [228]
P._fremontii           TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [234]
P._balsamifera_2       TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [233]
P._balsamifera_3       TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [233]
P._balsamifera_1       TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [233]
P._tremuloides_2       TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [194]
P._nigra_3             TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [196]
P._laurifolia_1        TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [230]
P._davidiana_1         TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [233]
P._grandidentata_2     TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [234]
P._grandidentata_3     TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [234]
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P._grandidentata_4    TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [233]
P._deltoides_1        TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCCTCCTTG   [229]
P._sargentii          TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCCTCCTTG   [228]
P._deltoides_2        TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCCTCCTTG   [229]
P._deltoides_3        TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCCTCCTTG   [230]
P._angulata           TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [233]
P._tremula_1          TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [238]
P._nigra_5            TCAAATGCTAGAATCAATCTACTCAATGATCGCTCTCGTCTTCATCCTTG   [233]

[                              260       270       280       290       300]
[                              .         .         .         .         .]

P._alba_1             TCACCTGCATGGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [288]
P._alba_3             TCACCTGCATGGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [288]
P._davidiana_4        TCACCTGCATGGAGCTCTGTGACGCAGCGACTGTCATCGACGTCTACCGC   [287]
P._trichocarpa_2      TCACCTTCATCGAGCTCTGTGACGCAACAACTGTCGTCGACGTCTACCGC   [207]
P._szechuanica        TCACCTGCATCGAGCTATGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [292]
p._maximowiczii_2     TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [284]
P._simonii            TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [283]
P._cathayana          TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [284]
P._maximowiczii_1     TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [285]
P._laurifolia_2       TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [284]
P._nigra_1            TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [283]
P._nigra_4            TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [284]
P._nigra_2            CCACGTGCATCGAGCTCTGTGACGCAACAACTGTCGTGGACGTCAACCGC   [278]
P._fremontii          TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [284]
P._balsamifera_2      TCACCTTCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [283]
P._balsamifera_3      TCACCTTCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [283]
P._balsamifera_1      TCACCTTCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [283]
P._tremuloides_2      TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [244]
P._nigra_3            TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [246]
P._laurifolia_1       TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [280]
P._davidiana_1        TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [283]
P._grandidentata_2    TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [284]
P._grandidentata_3    TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [284]
P._grandidentata_4    TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [283]
P._deltoides_1        TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [279]
P._sargentii          TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [278]
P._deltoides_2        TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [279]
P._deltoides_3        TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [280]
P._angulata           TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [283]
P._tremula_1          TCACCTGCATGGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [288]
P._nigra_5            TCACCTGCATCGAGCTCTGTGACGCAGCAACTGTCGTCGACGTCTACCGC   [283]

[                              310       320       330       340       350]
[                              .         .         .         .         .]

P._alba_1             TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [338]
P._alba_3             TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [338]
P._davidiana_4        TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTTCCAC   [337]
P._trichocarpa_2      TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCTCGTTTTGCCAC   [257]
P._szechuanica        TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [342]
p._maximowiczii_2     TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [334]
P._simonii            TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [333]
P._cathayana          TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [334]
P._maximowiczii_1     TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [335]
P._laurifolia_2       TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [334]
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P._nigra_1              TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [333]
P._nigra_4              TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [334]
P._nigra_2              TTAATCCAATACGATATCTCCGGAGCTGAGTTCGGATCGCGTTTTGCCAC   [328]
P._fremontii            TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [334]
P._balsamifera_2        TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCTCGTTTTGCCAC   [333]
P._balsamifera_3        TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCTCGTTTTGCCAC   [333]
P._balsamifera_1        TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCTCGTTTTGCCAC   [333]
P._tremuloides_2        TTAATCCAATTCGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [294]
P._nigra_3              TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [296]
P._laurifolia_1         TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [330]
P._davidiana_1          TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [333]
P._grandidentata_2      TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [334]
P._grandidentata_3      TTAATCCAATACGATATCTCCGGAGCTCATTTCGGATCGCGTTTTGCTAC   [334]
P._grandidentata_4      TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [333]
P._deltoides_1          TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [329]
P._sargentii            TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [328]
P._deltoides_2          TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [329]
P._deltoides_3          TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [330]
P._angulata             TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [333]
P._tremula_1            TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [338]
P._nigra_5              TTAATCCAATACGATATCTCCGGAGCTCAGTTCGGATCGCGTTTTGCCAC   [333]

[                              360       370       380       390       400]
[                              .         .         .         .         .]

P._alba_1               TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [388]
P._alba_3               TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [388]
P._davidiana_4          TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [387]
P._trichocarpa_2        TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [307]
P._szechuanica          TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [392]
p._maximowiczii_2       TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [384]
P._simonii              TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [383]
P._cathayana            TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [384]
P._maximowiczii_1       TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGCCGATCTTT   [385]
P._laurifolia_2         TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [384]
P._nigra_1              TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [383]
P._nigra_4              TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [384]
P._nigra_2              TTTGAACCACCACGCCGGCTCCGTGCATCTCCCTCCCGGCGTCGATCTTA   [378]
P._fremontii            TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [384]
P._balsamifera_2        TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [383]
P._balsamifera_3        TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [383]
P._balsamifera_1        TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [383]
P._tremuloides_2        TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [344]
P._nigra_3              TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [346]
P._laurifolia_1         TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [380]
P._davidiana_1          TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [383]
P._grandidentata_2      TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [384]
P._grandidentata_3      TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [384]
P._grandidentata_4      TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [383]
P._deltoides_1          TTTGAACCACCACGCCGGCTCCTTGCATCTTCCTCCCGGCGTCGATCTTT   [379]
P._sargentii            TTTGAACCACCACGCCGGCTCCTTGCATCTTCCTCCCGGCGTCGATCTTT   [378]
P._deltoides_2          TTTGAACCACCACGCCGGCTCCTTGCATCTTCCTCCCGGCGTCGATCTTT   [379]
P._deltoides_3          TTTGAACCACCACGCCGGCTCCTTGCATCTTCCTCCCGGCGTCGATCTTT   [380]
P._angulata             TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [383]
P._tremula_1            TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [388]
P._nigra_5              TTTGAACCACCACGCCGGCTCCTTGCATCTCCCTCCCGGCGTCGATCTTT   [383]
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[                          410        420        430        440        450]
[                          .          .          .          .          .]

P._alba_1                CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACTTTGGTT   [438]
P._alba_3                CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACTTTGGTT   [438]
P._davidiana_4           CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACTTTGGTT   [437]
P._trichocarpa_2         CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACGTTGGTT   [357]
P._szechuanica           CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACTTTGGTT   [442]
p._maximowiczii_2        CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACGTTGGTT   [434]
P._simonii               CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACGTTGGTT   [433]
P._cathayana             CTCGCACTGTCGTCATCGTTCCTGTTCGTGAATTGAATATCACGTTGGTT   [434]
P._maximowiczii_1        CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACGTTGGTT   [435]
P._laurifolia_2          CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACGTTGGTT   [434]
P._nigra_1               CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACGTTGGTT   [433]
P._nigra_4               CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACGTTGGTT   [434]
P._nigra_2               CTCGTACTGTCGTGATCATTCCTGTTCGGGAATTGAATATCACGTCGGTT   [428]
P._fremontii             CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACGTTGGTT   [434]
P._balsamifera_2         CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACGTTGGTT   [433]
P._balsamifera_3         CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACGTTGGTT   [433]
P._balsamifera_1         CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACGTTGGTT   [433]
P._tremuloides_2         CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACTTTGATT   [394]
P._nigra_3               CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACGTTGGTT   [396]
P._laurifolia_1          CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACGTTGGTT   [430]
P._davidiana_1           CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACTTTGATT   [433]
P._grandidentata_2       CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACTTTGATT   [434]
P._grandidentata_3       CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACTTTGATT   [434]
P._grandidentata_4       CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACTTTGATT   [433]
P._deltoides_1           CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACTTTGGTT   [429]
P._sargentii             CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACTTTGGTT   [428]
P._deltoides_2           CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACTTTGGTT   [429]
P._deltoides_3           CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACTTTGGTT   [430]
P._angulata              CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACGTTGGTT   [433]
P._tremula_1             CTCGCACTGTTGTCATCATTCCTGTTCGTGAATTGAATATCACTTTGGTT   [438]
P._nigra_5               CTCGCACTGTCGTCATCATTCCTGTTCGTGAATTGAATATCACGTTGGTT   [433]

[                          460        470        480        490        500]
[                          .          .          .          .          .]

P._alba_1                AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATAACAGCCTA   [488]
P._alba_3                AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATAACAGCCTA   [488]
P._davidiana_4           AAAGGTTAATTACATTTCTGATTGCGCATCTCGTCTATAATGACAGCCTA   [487]
P._trichocarpa_2         AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATGACAGCCTA   [407]
P._szechuanica           AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATGACAGCCTA   [492]
p._maximowiczii_2        AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATGACAGCCTA   [484]
P._simonii               AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATGACAGCCTA   [483]
P._cathayana             AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATGACAGCCTA   [484]
P._maximowiczii_1        AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATGACAGCCTA   [485]
P._laurifolia_2          AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATGACAGCCTA   [484]
P._nigra_1               AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATGACAGCCTA   [483]
P._nigra_4               AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATGACAGCCTA   [484]
P._nigra_2               AAAGGATAAGTACATTTCTAATTGCGCAGCTAGTCTATAATAACAGGCTA   [478]
P._fremontii             AAAGGTTAATTACATTTCTAATTGCGTATCTAGTCTATAATAACAGCCTA   [484]
P._balsamifera_2         AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATGACAGCCTA   [483]
P._balsamifera_3         AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATGACAGCCTA   [483]
P._balsamifera_1         AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATGACAGCCTA   [483]
P._tremuloides_2         AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATGACAGCCTA   [444]
P._nigra_3               AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATGACAGCCTA   [446]
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```
P._laurifolia_1        AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATGACAGCCTA   [480]
P._davidiana_1         AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATGACAGCCTA   [483]
P._grandidentata_2     AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATGACAGCCTA   [484]
P._grandidentata_3     AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATGACAGCCTA   [484]
P._grandidentata_4     AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATGACAGCCTA   [483]
P._deltoides_1         AAAGGTTAATTACATTTCTAATTGCGTATCTAGTATATAATGACAGCCTA   [479]
P._sargentii           AAAGGTTAATTACATTTCTAATTGCGTATCTAGTATATAATGACAGCCTA   [478]
P._deltoides_2         AAAGGTTAATTACATTTCTAATTGCGTATCTAGTATATAATGACAGCCTA   [479]
P._deltoides_3         AAAGGTTAATTACATTTCTAATTGCGTATCTAGTATATAATGACAGCCTA   [480]
P._angulata            AAAGGTTAATTACATTTCTAATTGCGTATCTAGTCTATAATAACAGCCTA   [483]
P._tremula_1           AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATTACAGCCTG   [488]
P._nigra_5             AAAGGTTAATTACATTTCTAATTGCGCATCTAGTCTATAATGACAGCCTA   [483]

[                              510        520        530        540        550]
[                              .          .          .          .          .]

P._alba_1              CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [537]
P._alba_3              CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [537]
P._davidiana_4         CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [536]
P._trichocarpa_2       CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTATTAGCTT   [457]
P._szechuanica         CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [541]
p._maximowiczii_2      CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [533]
P._simonii             CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [532]
P._cathayana           CTTACTTAAAATTTGGTTAAGTAATTAAATTGGCTGGTTACTAT-AGCTT   [533]
P._maximowiczii_1      CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [534]
P._laurifolia_2        CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [533]
P._nigra_1             CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [532]
P._nigra_4             CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [533]
P._nigra_2             CTTACTTAACATTTGGTTAAGTAATTAAATTGGATGGTTACTAT-AGCTT   [527]
P._fremontii           CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [533]
P._balsamifera_2       CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [532]
P._balsamifera_3       CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [532]
P._balsamifera_1       CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [532]
P._tremuloides_2       CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [493]
P._nigra_3             CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [495]
P._laurifolia_1        CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [529]
P._davidiana_1         CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [532]
P._grandidentata_2     CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [533]
P._grandidentata_3     CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [533]
P._grandidentata_4     CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [532]
P._deltoides_1         CTTACTTAAAATTTGGTTATGTAATTAAATTGGTTGGTTACTGT-AGCTT   [528]
P._sargentii           CTTACTTAAAATTTGGTTATGTAATTAAATTGGTTGGTTACTGT-AGCTT   [527]
P._deltoides_2         CTTACTTAAAATTTGGTTATGTAATTAAATTGGTTGGTTACTGT-AGCTT   [528]
P._deltoides_3         CTTACTTAAAATTTGGTTATGTAATTAAATTGGTTGGTTACTGT-AGCTT   [529]
P._angulata            CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [532]
P._tremula_1           CTTACTTAAAATTTGGCTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [537]
P._nigra_5             CTTACTTAAAATTTGGTTAAGTAATTAAATTGGTTGGTTACTAT-AGCTT   [532]

[                              560        570        580        590        600]
[                              .          .          .          .          .]

P._alba_1              AATTAATTAAAGTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [587]
P._alba_3              AATTAATTAAAGTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [587]
P._davidiana_4         AATTAATTAAAGTAAAATGTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [586]
P._trichocarpa_2       AAGTAATTT-------------------------------------------   [466]
P._szechuanica         AAGTAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [590]
p._maximowiczii_2      AATTAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [582]
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P._simonii              AATTAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [581]
P._cathayana            AATCAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [582]
P._maximowiczii_1       AATTAATGAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [583]
P._laurifolia_2         AATTAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [582]
P._nigra_1              AATTAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [581]
P._nigra_4              AATTAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [582]
P._nigra_2              AATTAATTAA-GTAAAATTTAATGAAGCGATTAAATTGTTTGATTAGAAT   [576]
P._fremontii            AATTAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [582]
P._balsamifera_2        AATCAATCAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [581]
P._balsamifera_3        AATCAATCAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [581]
P._balsamifera_1        AATCAATCAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [581]
P._tremuloides_2        AATTAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [542]
P._nigra_3              AATTAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [544]
P._laurifolia_1         AATTAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [578]
P._davidiana_1          AATTAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [581]
P._grandidentata_2      AATTAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [582]
P._grandidentata_3      AATTAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [582]
P._grandidentata_4      AATTAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [581]
P._deltoides_1          AATTAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [577]
P._sargentii            AATTAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [576]
P._deltoides_2          AATTAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [577]
P._deltoides_3          AATTAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [578]
P._angulata             AATTAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [581]
P._tremula_1            AATTAATTAA----------------------------------------   [547]
P._nigra_5              AATTAATTAA-GTAAAATTTAATGAAGCAGTTAAATTGTTTGATTAGTAT   [581]

[                                610       620       630       640       650]
[                                .         .         .         .         .]

P._alba_1               GGATTAATTAATTAAAATTTAGTGAAGCAGTTAAATCAGTTGATTTTTAT   [637]
P._alba_3               GGATTAATTAATTAAAATTTA----------------------------   [608]
P._davidiana_4          GGATTAATTAATTAAAATTTAGTGAAGCAGTTAAATCAGTTGATTTTTAT   [636]
P._trichocarpa_2        --------------------------------------------------   [466]
P._szechuanica          AGCTTAATTAATTAAAATTTAATGAAGTAGTTAAATCAGTTGATTTTTAT   [640]
p._maximowiczii_2       AGCTTAATTAATTAAAATTTAATGAAGTAGTTAAATCAGTTGATTTTTGT   [632]
P._simonii              AGCTTAATTAATTAAAATTTAATGAAGTAGTTAAATCAGTTGATTTTTGT   [631]
P._cathayana            AGCTTAATTAATTAAAATTTAATGAAGTAGTTAAATCAGTTGATTTTTAT   [632]
P._maximowiczii_1       AGCTTAATTAATTAAAATTTA-TGAAGTAGTTAAATCAGTTGATTTTTAT   [632]
P._laurifolia_2         AGCTTAATTAATTAAAATTTA-TGAAGTAGTTAAATCAGTTGATTTTTAT   [631]
P._nigra_1              AGCTTAATTAATTAAAATTTAATGAAGTAGTTAAATCAGTTGATTTTTAT   [631]
P._nigra_4              AGCTTAATTAATTAAAATTTAATGAAGTAGTTAAATCAGTTGATTTTTAT   [632]
P._nigra_2              AGCTTAATTAATTAAAATTTAATGAAGTGGTTAAATCAGATGATTTGAAT   [626]
P._fremontii            AGCTTAATTAATTAAAATTTAATGAAGTAGTTAAATCAGTTGATTTTTAT   [632]
P._balsamifera_2        AGCTTAATTAATTAAAATTTAATGAAGTAGTTAAATCAGTTGATTTTTAT   [631]
P._balsamifera_3        AGCTTAATTAATTAAAATTTAATGAAGTAGTTAAATCAGTTGATTTTTAT   [631]
P._balsamifera_1        AGCTTAATTAATTAAAATTTAATGAAGTAGTTAAATCAGTTGATTTTTAT   [631]
P._tremuloides_2        AGCTTAATTAATTAAAATTTAATGAGGCAGTTAAATCAGTTGATTTTTAT   [592]
P._nigra_3              AGCTTAATTAATTAAAATTTAATGAAGTAGTTAAATCAGTTGATTTTTAT   [594]
P._laurifolia_1         AGCTTAATTAATTAAAATTTA-TGAAGTAGTTAAATCAGTTGATTTTTAT   [627]
P._davidiana_1          AGCTTAATTAATTAAAATTTAATGAGGCAGTTAAATCAGTTGATTTTTAT   [631]
P._grandidentata_2      AGCTTAATTAATTAAAATTTAATGAGGCAGTTAAATCAGTTGATTTTTAT   [632]
P._grandidentata_3      AGCTTAATTAATTAAAATTTAATGAGGCAGTTAAATCAGTTGATTTTTAT   [632]
P._grandidentata_4      AGCTTAATTAATTAAAATTTAATGAGGCAGTTAAATCAGTTGATTTTTAT   [631]
P._deltoides_1          AGCTTAATTAATTAAAATTTAATGAAGCAGTTAAATCAGTTGATTTTTAT   [627]
P._sargentii            AGCTTAATTAATTAAAATTTAATGAAGCAGTTAAATCAGTTGATTTTTAT   [626]
P._deltoides_2          AGCTTAATTAATTAAAATTTAATGAAGCAGTTAAATCAGTTGATTTTTAT   [627]
P._deltoides_3          AGCTTAATTAATTAAAATTTAATGAAGCAGTTAAATCAGTTGATTTTTAT   [628]
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P._angulata          AGCTTAATTAATTAAAATTTAATGAAGTAGTTAAATCAGTTGATTTTTAT   [631]
P._tremula_1         AG----------TAAAATTTAATGAAGCAGTTAAATCAGTTGATTTTTAT   [587]
P._nigra_5           AGCTTAATTAATTAAAATTTAATGAAGTAGTTAAATCAGTTGATTTTTAT   [631]

[                              660       670       680       690       700]
[                              .         .         .         .         .]

P._alba_1            GAATTTTTATGATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [687]
P._alba_3            ----------------------------------------------------   [608]
P._davidiana_4       GAT----------AGAGTTTATTGCGCAGAGAAAACCGCTGGGAGGATTA   [676]
P._trichocarpa_2     ----------------------------------------------------   [466]
P._szechuanica       GATTTTTTATGTTAGAGTTTATTGCACAAAGAAAACCGCTGGGAGGATTA   [690]
p._maximowiczii_2    GATTTTTTATGATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [682]
P._simonii           GATTTTTTATGATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [681]
P._cathayana         GATTTTTTATGATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [682]
P._maximowiczii_1    GATTTTTTATG-TAGAGTTTATCGCGCAAAGAAAACCGCTGGGAGGATTA   [681]
P._laurifolia_2      GATTTTTTATG-TAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [680]
P._nigra_1           GATTTTTTATGATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [681]
P._nigra_4           GATTTTTTATGATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [682]
P._nigra_2           GATATGATATGATAGAGTAGATTGCGCAGAGAAAACCGCTGGGA------   [670]
P._fremontii         GATTTTTTATGATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [682]
P._balsamifera_2     GATTTTTTATGATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [681]
P._balsamifera_3     GATTTTTTATGATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [681]
P._balsamifera_1     GATTTTTTATGATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [681]
P._tremuloides_2     GATTTTTTATTATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [642]
P._nigra_3           GATTTTTTATGATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [644]
P._laurifolia_1      GATTTTTTATG-TAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [676]
P._davidiana_1       GATTTTTTATTATAGAGTTTATTGTGCAAAGAAAACCGCTGGGAGGATTA   [681]
P._grandidentata_2   GATTTTTTATTATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [682]
P._grandidentata_3   GATTTTTTATTATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [682]
P._grandidentata_4   GATTTTTTATTATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [681]
P._deltoides_1       GATTTTTTATCATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [677]
P._sargentii         GATTTTTTATCATAGAGTTTATTGCGCAAAGA------------------   [658]
P._deltoides_2       GATTTTTTATCATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [677]
P._deltoides_3       GATTTTTTATCATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [678]
P._angulata          GATTTTTTATGATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [681]
P._tremula_1         GAATTTTTATGATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [637]
P._nigra_5           GATTTTTTATGATAGAGTTTATTGCGCAAAGAAAACCGCTGGGAGGATTA   [681]

[                              710       720       730       740       750]
[                              .         .         .         .         .]

P._alba_1            TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [737]
P._alba_3            ----------------------------------------------------   [608]
P._davidiana_4       TTATATTTGCT--------------------------------------   [687]
P._trichocarpa_2     ----------------------------------------------------   [466]
P._szechuanica       TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [740]
p._maximowiczii_2    TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [732]
P._simonii           TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [731]
P._cathayana         TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [732]
P._maximowiczii_1    TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAACAGAGACCCCACGTC   [731]
P._laurifolia_2      TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [730]
P._nigra_1           TTATTTTTGCTGCCGC----------------------------------   [697]
P._nigra_4           TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [732]
P._nigra_2           ----------------------------------------------------   [670]
P._fremontii         TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [732]
P._balsamifera_2     TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [731]
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P._balsamifera_3      TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [731]
P._balsamifera_1      TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [731]
P._tremuloides_2      TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [692]
P._nigra_3            TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [694]
P._laurifolia_1       TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [726]
P._davidiana_1        TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [731]
P._grandidentata_2    TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [732]
P._grandidentata_3    TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [732]
P._grandidentata_4    TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [731]
P._deltoides_1        TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [727]
P._sargentii          --------------------------------------------------   [658]
P._deltoides_2        TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [727]
P._deltoides_3        TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [728]
P._angulata           TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [731]
P._tremula_1          TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [687]
P._nigra_5            TTATTTTTGCTGCCGCAAATGCTTAATTTTGAAAATAGAGACCCCACGTC   [731]

[                              760       770       780       790       800]
[                              .         .         .         .         .]

P._alba_1             TGAGAGCAAGTATCAAATACATGAGAAGAAGTTGACAAAGAATGTTTTGG   [787]
P._alba_3             --------------------------------------------------   [608]
P._davidiana_4        --------------------------------------------------   [687]
P._trichocarpa_2      --------------------------------------------------   [466]
P._szechuanica        TGAGAGCAAGTATCAAATACATGAGAAGAAGTTGACAAAG----------   [780]
p._maximowiczii_2     TGAGAGCAAGTATCANATACATGAGAAGAAGTTGACAAACAATGTTTTGG   [782]
P._simonii            TGAGAGCAAGTATCAAATACATGAGAAGAAGTTGACAAACAATGTTTTGG   [781]
P._cathayana          TGAGAGCAAGTATCAAATACATGAGAAGAAGTTGACAAACAATGTTTTGG   [782]
P._maximowiczii_1     TGAGAGCAAGTATCANATACATGAGAAGAAGTTGACAAACAATGTTTTGG   [781]
P._laurifolia_2       TGAGAGCAAGTATCAAATACATGAGAAGAAGTTGACAAACAATGTTTTGG   [780]
P._nigra_1            --------------------------------------------------   [697]
P._nigra_4            TGAGAGCAAGTTCCAAATACATGAGAAGAAGTTGACAAACAATGTTTTGG   [782]
P._nigra_2            --------------------------------------------------   [670]
P._fremontii          TGAGAGCAAATATC------------------------------------   [746]
P._balsamifera_2      TGAGAGCAAGTATCAAATACATGAAAAGAAGTTGACAAACAATGTTTTGG   [781]
P._balsamifera_3      TGAGAGCAAGTATCAAATACATGAAAAGAAGTTGACAAACAATGTTTTGG   [781]
P._balsamifera_1      TGAGAGCAAGTATCANATACATGAAAAGAAGTTGACAAACAATGTTTTGG   [781]
P._tremuloides_2      TGAGAGCAAGTATCAAATACATGAGAAGAAGTTGACAAAGAATGTTTTGG   [742]
P._nigra_3            TGAGAGCAAGTTCCAAATACATGAGAAGAAGTTGACAAACAATGTTTTGG   [744]
P._laurifolia_1       TGAGAGCAAGTATCAAATACATGAGAAGAAGTTGACAAACAATGTTTTGG   [776]
P._davidiana_1        TGAGAGCAAGTATCAAATACATGAGAAGAAGTTGACAAAGAATGTTTTGG   [781]
P._grandidentata_2    TGAGAGCAAGTATCAAATACATGAGAAGAAGTTGACAAAGAATGTTTTGG   [782]
P._grandidentata_3    TGAGAGCAAGTATCAAATACATGAG------------------------   [757]
P._grandidentata_4    TGAGAGCAAGTATCAAATACATGAGAAGAAGTTGACAAAGAATGTTTTGG   [781]
P._deltoides_1        TGAGAGCAAGTATCAAATACATGAGAAGAAGTTGACAAAGAATGTTTTGG   [777]
P._sargentii          --------------------------------------------------   [658]
P._deltoides_2        TGAGAGCAAGTATCAAATACATGAGAAGAAGTTGACAAAGAATGTTTTGG   [777]
P._deltoides_3        TGAGAGCAAGTATCAAATACATGAGAAGAAGTTGACAAAGAATGTTTTGG   [778]
P._angulata           TGAGAGCAAATATCAAATACATGAGAAGAAGTTGACAAACAATGTTTTGG   [781]
P._tremula_1          TGAGAGCAAGTATCAAATACATGAGAAGAAGTTGACAAAGAATGTTTTGG   [737]
P._nigra_5            TGAGAGCAAGTTCCAAATACATGAGAAGAAGTTGACAAACAATGTTTTGG   [781]

[                              810       820       830       840  ]
[                              .         .         .         .    ]

P._alba_1             TGGAGCTTGAACGATTGCTTATT----------------------   [810]
P._alba_3             ---------------------------------------------   [608]
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P._davidiana_4          ----------------------------------------------  [687]
P._trichocarpa_2        ----------------------------------------------  [466]
P._szechuanica          ----------------------------------------------  [780]
p._maximowiczii_2       TGGAGCTTGAACGATTGCTTATTTATGCCAATATTCCCG-------  [821]
P._simonii              TGGAGCTTGAACGATTGCTTATTTATGCCAATATTCCCGTGAGT    [825]
P._cathayana            TGGAGCTTGAACGATTGCTTATTTATGCCAATATTCCCGTGAGT    [826]
P._maximowiczii_1       TGGAGCTTGAACGATTGCTTATTTATGCCAATATTCCCGTGAGT    [825]
P._laurifolia_2         TGGAGCTTGAACGATTGCTTATTTATGCCAATATTCCCGTGAGT    [824]
P._nigra_1              ----------------------------------------------  [697]
P._nigra_4              TGGAGCTTGAACGATTGCTTATTTATG------------------    [809]
P._nigra_2              ----------------------------------------------  [670]
P._fremontii            ----------------------------------------------  [746]
P._balsamifera_2        TGGAGCTTGAACGATTGCTTATTTATGCCAATATTC----------  [817]
P._balsamifera_3        T---------------------------------------------  [782]
P._balsamifera_1        TGGAGCTTGAACGATTGCTTATTTATGCCAATATTCCCGTGAGT    [825]
P._tremuloides_2        TGGAGCTTGAACGATTGCTTATTTATGCCAATATTCCCGTGAGT    [786]
P._nigra_3              TGGAGCTTGAACGATTGCTTATTTATGCCAATATTCCCGTGAGT    [788]
P._laurifolia_1         TGGAGCTTGAACGATTGCTTATTTATGCCAATATTCCCGTGAGT    [820]
P._davidiana_1          TGGAGCTTGAACGATTGCTTATTTATGCCAATATTCCCGTGAGT    [825]
P._grandidentata_2      TGGAGCTTGAACGATTGCTTATTTATGCCAATATTCCCGTGAGT    [826]
P._grandidentata_3      ----------------------------------------------  [757]
P._grandidentata_4      TGGAGCTTG-------------------------------------  [790]
P._deltoides_1          TGGAGCTTGAACGATTGCTTATTTATGCCAATATTCCCGTGAGT    [821]
P._sargentii            ----------------------------------------------  [658]
P._deltoides_2          TGGAGCTTGAACGATTGCTTATTTATGCCAATATTCCCGTGAGT    [821]
P._deltoides_3          TGGAGCTTGAACGATTGCTTATTTATGCCAATATTCCCGTGAGT    [822]
P._angulata             TGGAGCTTGAACGATTGCTTATTTATGCCAATATTCCCGTGAGT    [825]
P._tremula_1            TGGAGCTTGAACGATTGCTTATTTATGCCAATATTCCCGTGAGT    [781]
P._nigra_5              TGGAGCTTGAACGATTGCTTATTTATGCCAATATTCCCGTGAGT    [825]
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Appendix 2. 4 Nucleotide sequence data matrix of SCISSR fragment PIS6A1.

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[                              10         20         30         40         50]
[                              .          .          .          .          .]

P._alba_1                CCCTTCATTACCTTCACAGAATCCTATGG-TCCATA-CTTCACAG-CAAA   [47]
P._alba_3                CCCTTCATTACCTTCACAGAATCCTATGG-TCCATA-CTTCACAG-CAAA   [47]
P._alba_2                CCCTTCATTACCTTCACAGAATCCTATGG-TCCATA-CTTCACAG-CAAA   [47]
P._grandidentata_2       CCCTTCATTACCTTCACAGAATCCTATGG-TCCATA-CTTCACAG-CAAA   [47]
P._grandidentata_3       CACTTCATTACCTTCACAGAATCCTATGG-TCCATA-CTTCACAG-CAAA   [47]
P._grandidentata_4       ---TTCATTACCTTCACAGAATCCTATGG-TCCATA-CTTCACAG-CAAA   [44]
P._davidiana_2           CCCTTCATTACCTTCACAGAATCCTATGG-TCCATA-CTTCACAG-CAAA   [47]
P._davidiana_5           ---TTCATTACCTTCACAGAATCCTATGG-TCCATA-CTTCACAG-CAAA   [44]
P._davidiana_4           -----------CTTCACAGAATCCTATGG-TCCATA-CTTCACAG-CAAA   [36]
P._davidiana_3           --CTTC-TTACCTTCACAGAATCCTATGG-TCCATA-CTTCACAG-CAAA   [44]
P._tremula_1             ---TTCATTACCTTCACAGAATCCTATGG-TCCATA-CTTCACAG-CAAA   [44]
P._tremuloides_2         CGCTTCATTACCTTCACAGAATCCTATGG-TCCATA-CTTCACAG-CAAA   [47]
P._davidiana_1           ---TTCATTACCTTCACAGAATCCTATGG-TCCATA-CTTCACAG-CAAA   [44]
P._tremuloides_1         --CTTCATTACCTTCACAGAATCCTATGG-TCCATA-CTTCACAG-CAAA   [45]
P._trichocarpa_1         CACTTCATTACCTTCACAGAATCCTCTGG-TCCATA-CTTCACAG-CAAA   [47]
P._balsamifera_1         CACTTCATTACCTTCACAGAATCCTCTGG-TCCATA-CTTCACAG-CAAA   [47]
P._balsamifera_2         -CCTTC-TTACCTTCACAGAATCCTCTGG-TCCATA-CTTCACAG-CAAA   [45]
P._cathayana             -----------CTTCACAGAATCCTCTGG-TCCATA-CTTCACAG-CAAA   [36]
P._tristis               ------------------AATCCTCTGG-TCCATA-CTTCACAG-CAAA   [28]
P._angustifolia          ---TTCATTACCTTCACAGAATCCTCTGG-TCCATA-CTTCACAG-CAAA   [44]
P._trichocarpa_2         -----------CTTCACAGAATCCTCTGG-TCCATA-CTTCACAG-CAAA   [36]
P._balsamifera_3         ---------ACCTT-CCAGAATCCTCTGG-TCCATA-CTTCACAG-CAAA   [37]
P._szechuanica           ---TTCATTACCTTCACAGAATCCTCTGG-TCCATA-CTTCACAG-CAAA   [44]
P._nigra_3               ---TTCATTACCTTCACAGAATCCTCTGG-TCCATA-CTTCACAG-CAAA   [44]
P._nigra_5               ---TTCATTACCTTCACAGAATCCTCTGG-TCCATA-CTTCACAG-CAAA   [44]
P._nigra_1               ---TTC-TTACCTTC-CAGAATCCTCTGG-TCCATA-CTTCACAG-CAAA   [42]
P._nigra_4               --CTTCATTACCTTCACAGAATCCTCTGG-TCCATA-CTTCACAG-CAAA   [45]
P._nigra_2               CACTTCATTACCTTCACAGAATCCTCTGG-TCCANA-CTTCACAG-CAAA   [47]
S._integra               CA-TTCATTACCTTCACAGAATCCTCTGGGTCCATGGCTTCACAG-CAAA   [48]
S._nigra                 CA-TTCATTACCTTCACAGAATCCTCTGG-TCCATA-CTTCACAG-CAAA   [46]
P._fremontii             ---TTCATTACCTTCACAGAATCCTCTGG-TCCATA-CTTCACAG-CAAA   [44]
P._angulata              ----------CCTTCACAGAATCCTCTGG-TCCATA-CTTCACAG-CAAA   [37]
P._laurifolia_1          CACTTCATTACCTTCACAGAATCCTCTGGGTCCATA-CTTCACAG-CAAA   [48]
P._laurifolia_2          ---TTCATTACCTTCACAGAATCCTCTGG-TCCATA-CTTCACAG-CAAA   [44]
P._simonii               ---TTCATTACCTTCACAGAATCCTCTGG-TCCATA-CTTCACAGGCAAA   [45]
P._maximowiczii_1        CACTTCATTACCTTCACAGAATCCTCTGG-TCCATA-CTTCACAG-CAAA   [47]
P._maximowiczii_2        CACTTCATTACCTTCACAGAATCCTCTGG-TCCATA-CTTCACAG-CAAA   [47]
P._deltoides_1           --------------------------------------------------   [0]
P._sargentii             ---TTCATTACCTTCACAGAATCCTCTGGGTCCATA-CTTCACAG-CAAA   [45]
P._deltoides_2           CACTTCATTACCTTCACAGAATCCTCTGG-TCCATA-CTTCACAG-CAAA   [47]
P._deltoides_3           CACTTCATTACCTTCACAGAATCCTCTGG-TCCATA-CTTCACAG-CAAA   [47]

[                              60         70         80         90        100]
[                              .          .          .          .          .]

P._alba_1                TCCAGG-TTCCGGGATCTGATAATTTAC-TTGCTTTTGGCCCTCGACAGA   [95]
P._alba_3                TCCAGG-TTCCGGGATCTGATAATTTAC-TTGCTTTTGGCCCTCGACAGA   [95]
P._alba_2                TCCAGG-TTCCGGGATCTGATAATTTAC-TTGCTTTTGGCCCTCGACAGA   [95]
P._grandidentata_2       TCCAGG-TTCCGGGATCTGATAATTTAC-TTGCTTTTGGCCCTCGACAGA   [95]
P._grandidentata_3       TCCAGG-TTCCGGGATCTGATAATTTAC-TTGCTTTTGGCCCTCGACAGA   [95]
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P._grandidentata_4    TCCAGG-TTCCGGGATCTGATAATTTAC-TTGCTTTTGGCCCTCGACAGA   [92]
P._davidiana_2        TCCAGG-TTCCGGGATCTGATAATTTAC-TTGCTTTTGGCCCTCGACAGA   [95]
P._davidiana_5        TCCAGG-TTCCGGGATCTGATAATTTAC-TTNCTTTTGGCCCTCGACAGA   [92]
P._davidiana_4        TCCAGG-TTCCGGGATCTGATAATTTAC-TTGCTTTTGGCCCTCGACAGA   [84]
P._davidiana_3        TCCAGG-TTCCGGGATCTGATAATTTAC-TTGCTTTTGGCCCTCCACAGA   [92]
P._tremula_1          TCCAGG-TTCCGGGATCTGATAATTTAC-TTGCTTTTGGCCCTCGACAGA   [92]
P._tremuloides_2      TCCAGG-TTCCGGGATCTGATAATTTAC-TTGCTTTTGGCCCTCGACAGA   [95]
P._davidiana_1        TCCAGG-TTCCGGGATCTGATAATTTAC-TTGCTTTTGGCCCTCCACAGA   [92]
P._tremuloides_1      TCCAGG-TTCCGGGATCTGATAATTTAC-TTGCTTTTGGCCCTCGACAGA   [93]
P._trichocarpa_1      TCCAGG-TTCCGGGATCTGACAATTTAC-TTGCTCTTGGCCGTCCACAGA   [95]
P._balsamifera_1      TCCAGG-TTCCGGGATCTGACAATTTAC-TTGCTCTTGGCCGTCCACAGA   [95]
P._balsamifera_2      TCCAGG-TTCCGGGATCTGACAATTTAC-TTGCTCTTGGCCGTCCACAGA   [93]
P._cathayana          TCCAGG-TTCCGGGATCTGACAATTTAC-TTGCTCTTGGCCGTCCACAGA   [84]
P._tristis            TCCAGG-TTCCGGGATCTGACAATTTAC-TTGCTCTTGGCCGTCCACAGA   [76]
P._angustifolia       TCCAGG-TTCCGGGATCTGACAATTTAC-TTGCTCTTGGCCGTCCACAGA   [92]
P._trichocarpa_2      TCCAGG-TTCCGGGATCTGACAATTTAC-TTGCTCTTGGCCGTCCACAGA   [84]
P._balsamifera_3      TCCAGG-TCCCGGGATCTGACAATTTAC-TTGCTCTTGGCCGTCCACAGA   [85]
P._szechuanica        TCCAGGGTTCCGGGATCTGACAATTTAC-TTGCTCTTGGCCGTCCACAGA   [93]
P._nigra_3            TCCAGG-TTCCAGGATCTGACAATTTAC-CTGCTCTTGGCCCTCCACAGA   [92]
P._nigra_5            TCCAGG-TTCCAGGATCTGACAATTTAC-CTGCTCTTGGCCCTCCACAGA   [92]
P._nigra_1            TCCAGG-TTCCAGGATCTGACAATTTAC-CTGCTCTTGGCCCTCCACAGA   [90]
P._nigra_4            TCCAGG-TTCCAGGATCTGACAATTTAC-CTGCTCTTGGCCCTCCACAGA   [93]
P._nigra_2            TCCAGG-TTCC-GGATCTGACAATTTAC-CTGCTCTTGGCCCTCCACAGA   [94]
S._integra            TCCAGGGTTCCGGGAACTGACAATTTAC-TTGTTCTTGCCCCTCCACAGA   [97]
S._nigra              TCCAGG-TTCCGGGAACTGACAATTTAC-TTGCTCTTGGCCCTCCACAGA   [94]
P._fremontii          TCCAGG-TTCCGGGATCTGACAATTTACCTTGCTCTTGGCCCTCCACAGA   [93]
P._angulata           TCCAGG-TTCCGGGATCTGACAATTTAC-TTGCTCTTGGCCCTCCACAGA   [85]
P._laurifolia_1       TCCAGG-TTCCGGGATCTGACAATTTAC-TTGCTCTTGGCCCTCCACAGA   [96]
P._laurifolia_2       TCCAGG-TTCCGGGATCTGACAATTTAC-TTGCTCTTGGCCCTCCACAGA   [92]
P._simonii            TCCAGGTT-CCGGGATCTGACAATTTAC-TTGCTCTTGGCCCTCCACAGA   [93]
P._maximowiczii_1     TCCAGG-TTCCGGGATCTGACAATTTAC-TTGCTCTTGGCCCTCCACAGA   [95]
P._maximowiczii_2     TCCAGG-TTCCGGGATCTGACAATTTAC-TTGCTCTTGGCCCTCCACAGA   [95]
P._deltoides_1        --------------------------------------------------   [0]
P._sargentii          TCCAGG--TTCCGGGATCTGACAATTTACTTGCTCTTGGCCCTCCACAGA   [93]
P._deltoides_2        TCCAGG--TTCCGGGATCTGACAATTTACTTGCTCTTGGCCCTCCACAGA   [95]
P._deltoides_3        TCCAGG--TTCCGGGATCTGACAATTTACTTGCTCTTGGCCCTCCACAGA   [95]

[                              110       120       130       140       150]
[                              .         .         .         .         .]

P._alba_1             CTTCTACACCTGGCTTTCAGAGTTTTGGCTCTTCGAATCTTAATTCATAC   [145]
P._alba_3             CTTCTACACCTGGCTTTCAGAGTTTTGGCTCTTCGAATCTTAATTCATAC   [145]
P._alba_2             CTTCTGCACCTGGCTTTCAGAGTTTTGGCTCTTCGAATCTTAATTCATAC   [145]
P._grandidentata_2    CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [145]
P._grandidentata_3    CTTCTACACCTGGCTTTCAGAGTTTTGGCTCCTCGAATCTTAATTCATAC   [145]
P._grandidentata_4    CTTCTACACCTGGCTTTCAGAGTTTTGGCTCCTCGAATCTTAATTCATAC   [142]
P._davidiana_2        CTTCTACACCAGGCTTTCAGAGTTTTGGCTCTTCGAATCTTAATTCATAC   [145]
P._davidiana_5        CTTCTACACCAGGCTTTCAGAGTTTTGGCTCTTCGAATCTTAATTCATAC   [142]
P._davidiana_4        CTTCTACACCAGGCTTTCAGAGTTTTGGCTCTTCGAATCTTAATTCATAC   [134]
P._davidiana_3        CTTCTACACCAGGCTTTCAGAGTTTTGGCTCTTCGAATCTTAATTCATAC   [142]
P._tremula_1          CTTCTACACCAGGCTTTCAGAGTTTTGGCTCTTCGAATCTTAATTCATAC   [142]
P._tremuloides_2      CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [145]
P._davidiana_1        CTTCTACACCAGGCTTTCAGAGTTTTGGCTCTTCGAATCTTAATTCATAC   [142]
P._tremuloides_1      CTTCTATACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [143]
P._trichocarpa_1      CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [145]
P._balsamifera_1      CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [145]
P._balsamifera_2      CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [143]
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P._cathayana          CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [134]
P._tristis            CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [126]
P._angustifolia       CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [142]
P._trichocarpa_2      CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [134]
P._balsamifera_3      CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [135]
P._szechuanica        CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [143]
P._nigra_3            CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [142]
P._nigra_5            CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [142]
P._nigra_1            CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [140]
P._nigra_4            CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [143]
P._nigra_2            CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [144]
S._integra            CCTGCACATCAGGCTTTCAGAGTTTTGGCATTTCAAATCTTAATTCACAC   [147]
S._nigra              CCTGTACATCAGGCTTTCAGAGTTTTGGCATTTCAAATCTTAATTCACAC   [144]
P._fremontii          CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [143]
P._angulata           CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [135]
P._laurifolia_1       CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [146]
P._laurifolia_2       CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [142]
P._simonii            CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [143]
P._maximowiczii_1     CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [145]
P._maximowiczii_2     CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [145]
P._deltoides_1        --------------------------------------------------   [0]
P._sargentii          CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [143]
P._deltoides_2        CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [145]
P._deltoides_3        CTTCTACACCAGGCTTTCAGAGTTTTGGCTCATCGAATCTTAATTCATAC   [145]

[                              160       170       180       190       200]
[                              .         .         .         .         .]

P._alba_1             AAGGGAACTGAGGATTTCTTCTCAGAAGACGAGATTCGTACGAGAAGTCA   [195]
P._alba_3             AAGGGAACTGAGGATTTCTTCTCAGAAGACGAGATTCGTACGAGAAGTCA   [195]
P._alba_2             AAGGGAACTGAGGATTTCTTCTCAGAAGACGAGATTCGTACGAGAAGTCA   [195]
P._grandidentata_2    AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [195]
P._grandidentata_3    AAGGGAACTGAGGATTTCTTCTCAGAAGANGAGATTCGTACGAGAAGTCA   [195]
P._grandidentata_4    AAGGGAACTGAGGATTTCTTCTCAGAAGANGAGATTCGTACGAGAAGTCA   [192]
P._davidiana_2        AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [195]
P._davidiana_5        AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [192]
P._davidiana_4        AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [184]
P._davidiana_3        AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [192]
P._tremula_1          AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [192]
P._tremuloides_2      AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACNAGAAGTCA   [195]
P._davidiana_1        AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACNAGAAGTCA   [192]
P._tremuloides_1      AAGGGAACTGANGATTTCTTCTCAGAAGAGGAGATTCGTACAAGAAGTCA   [193]
P._trichocarpa_1      AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [195]
P._balsamifera_1      AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [195]
P._balsamifera_2      AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [193]
P._cathayana          AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [184]
P._tristis            AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [176]
P._angustifolia       AAGGGAACTGAGGATTTTTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [192]
P._trichocarpa_2      AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [184]
P._balsamifera_3      AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [185]
P._szechuanica        AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [193]
P._nigra_3            AAGGGAATTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [192]
P._nigra_5            AAGGGAATTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [192]
P._nigra_1            AAGGGAATTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [190]
P._nigra_4            AAGGGAATTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [193]
P._nigra_2            AAGGGAANTGAGGATTTCTTCNCAGAAGAGGAGATTCGTACGAGAAGTCA   [194]
S._integra            AATGGAACTGAGGACTTCTTCTCAGAAGAGGAGATTCGTACAAGAAGTCA   [197]
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S._nigra              AACGGAACTGAGGACTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [194]
P._fremontii          AAGGGAANTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [193]
P._angulata           AAGGGAATTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [185]
P._laurifolia_1       AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [196]
P._laurifolia_2       AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [192]
P._simonii            AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [193]
P._maximowiczii_1     AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [195]
P._maximowiczii_2     AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [195]
P._deltoides_1        --------------------------------------------------   [0]
P._sargentii          AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [193]
P._deltoides_2        AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [195]
P._deltoides_3        AAGGGAACTGAGGATTTCTTCTCAGAAGAGGAGATTCGTACGAGAAGTCA   [195]

[                              210       220       230       240       250]
[                              .         .         .         .         .]

P._alba_1             TGAGATGCTTGAAAATGATGATATGCAGCATCTTCTTCGCATATTTAACA   [245]
P._alba_3             TGAGATGCTTGAAAATGATGATATGCAGCATCTTCTTCGCATATTTAACA   [245]
P._alba_2             TGAGATGCTTGAAAATGATGATATGCAGCATCTTCTTCGCATATTTAACA   [245]
P._grandidentata_2    TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [245]
P._grandidentata_3    TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [245]
P._grandidentata_4    TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [242]
P._davidiana_2        TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [245]
P._davidiana_5        TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [242]
P._davidiana_4        TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [234]
P._davidiana_3        TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [242]
P._tremula_1          TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [242]
P._tremuloides_2      TGAGATGCTTGAAAATGACGATATGCAGNATCTTCTTCGCATATTTAACA   [245]
P._davidiana_1        TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [242]
P._tremuloides_1      TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [243]
P._trichocarpa_1      TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [245]
P._balsamifera_1      TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [245]
P._balsamifera_2      TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [243]
P._cathayana          TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [234]
P._tristis            TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [226]
P._angustifolia       TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [242]
P._trichocarpa_2      TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [234]
P._balsamifera_3      TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [235]
P._szechuanica        TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [243]
P._nigra_3            TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [242]
P._nigra_5            TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [242]
P._nigra_1            TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [240]
P._nigra_4            TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [243]
P._nigra_2            TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [244]
S._integra            TGAGATGCTCGAAAACGACGATATGCAGCATCTTCTTCGCATATTCAACA   [247]
S._nigra              NGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCACATATTNAACA   [244]
P._fremontii          TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [243]
P._angulata           TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [235]
P._laurifolia_1       TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [246]
P._laurifolia_2       TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [242]
P._simonii            TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTNCNCATATTTAACA   [243]
P._maximowiczii_1     TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [245]
P._maximowiczii_2     TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [245]
P._deltoides_1        ----------------------------------------CATATTTAACA   [11]
P._sargentii          TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [243]
P._deltoides_2        TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [245]
P._deltoides_3        TGAGATGCTTGAAAATGACGATATGCAGCATCTTCTTCGCATATTTAACA   [245]
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[                          260        270        280        290        300]
[                          .          .          .          .          .]

P._alba_1                TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCANCGAAGATGGGTACCCT   [295]
P._alba_3                TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [295]
P._alba_2                TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [295]
P._grandidentata_2       TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [295]
P._grandidentata_3       TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [295]
P._grandidentata_4       TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [292]
P._davidiana_2           TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [295]
P._davidiana_5           TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [292]
P._davidiana_4           TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [284]
P._davidiana_3           TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [292]
P._tremula_1             TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [292]
P._tremuloides_2         TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [295]
P._davidiana_1           TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [292]
P._tremuloides_1         TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [293]
P._trichocarpa_1         TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [295]
P._balsamifera_1         TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [295]
P._balsamifera_2         TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [293]
P._cathayana             TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [284]
P._tristis               TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [276]
P._angustifolia          TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [292]
P._trichocarpa_2         TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [284]
P._balsamifera_3         TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [285]
P._szechuanica           TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [293]
P._nigra_3               TGGGAGGCCAGGGTCTAGCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [292]
P._nigra_5               TGGGAGGCCAGGGTCTAGCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [292]
P._nigra_1               TGGGAGGCCAGGGTCTAGCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [290]
P._nigra_4               TGGGAGGCCAGGGTCTAGCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [293]
P._nigra_2               TGGGAGGCCAGGGTCTAGCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [294]
S._integra               TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCCCTGAAGATGGGTACCCT   [297]
S._nigra                 TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCCCTGAAGATGGGTACCCT   [294]
P._fremontii             TGGGAGGCCAGGGTCTNTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [293]
P._angulata              TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [285]
P._laurifolia_1          TGGGAGGCCAGGGTCTAGCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [296]
P._laurifolia_2          TGGGAGGCCAGGGTCTAGCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [292]
P._simonii               TGGGAGGCCAGGGTCTAGCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [293]
P._maximowiczii_1        TGGGAGGCCAGGGTCTAGCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [295]
P._maximowiczii_2        TGGGAGGCCAGGGTCTAGCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [295]
P._deltoides_1           TGGGAGGCCANGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [61]
P._sargentii             TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [293]
P._deltoides_2           TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [295]
P._deltoides_3           TGGGAGGCCAGGGTCTCTCCTCCTTTAATGCCACCGAAGATGGGTACCCT   [295]

[                          310        320        330        340        350]
[                          .          .          .          .          .]

P._alba_1                TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [345]
P._alba_3                TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [345]
P._alba_2                TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [345]
P._grandidentata_2       TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [345]
P._grandidentata_3       TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [345]
P._grandidentata_4       TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [342]
P._davidiana_2           TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [345]
P._davidiana_5           TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [342]
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P._davidiana_4        TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [334]
P._davidiana_3        TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [342]
P._tremula_1          TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [342]
P._tremuloides_2      TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [345]
P._davidiana_1        TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [342]
P._tremuloides_1      TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [343]
P._trichocarpa_1      TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [345]
P._balsamifera_1      TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [345]
P._balsamifera_2      TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [343]
P._cathayana          TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [334]
P._tristis            TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [326]
P._angustifolia       TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [342]
P._trichocarpa_2      TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [334]
P._balsamifera_3      TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [335]
P._szechuanica        TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [343]
P._nigra_3            TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [342]
P._nigra_5            TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [342]
P._nigra_1            TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [340]
P._nigra_4            TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [343]
P._nigra_2            TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [344]
S._integra            TTCTCGTCAACAAACATACCTGCCGCATCTCCAAACTACTGCTTTGGTGA   [347]
S._nigra              TTNTCNTCATCAAACATACCTGCCGCATCTCCAAATTACTGCTTTGGTGA   [344]
P._fremontii          TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTNCAGCTTTGGTGA   [343]
P._angulata           TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTGCAGCTTTGGTGA   [335]
P._laurifolia_1       TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [346]
P._laurifolia_2       TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [342]
P._simonii            TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [343]
P._maximowiczii_1     TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [345]
P._maximowiczii_2     TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTACAGCTTTGGTGA   [345]
P._deltoides_1        TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTGCAGCTTTGTTGA   [111]
P._sargentii          TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTGCAGCTTTGTTGA   [343]
P._deltoides_2        TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTGCAGCTTTGTTGA   [345]
P._deltoides_3        TTCTCGTCAACAAACATGCCTACCGCACCTCCAAATTGCAGCTTTGTTGA   [345]

[                              360        370        380        390        400]
[                              .          .          .          .          .]

P._alba_1             TGATCCATCTNGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [395]
P._alba_3             TGATCCATCTTGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [395]
P._alba_2             TGATCCATCTTGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [395]
P._grandidentata_2    CGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [395]
P._grandidentata_3    CGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [395]
P._grandidentata_4    CGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [392]
P._davidiana_2        CGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [395]
P._davidiana_5        CGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [392]
P._davidiana_4        CGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGGTGGCTCAAGCTCA   [384]
P._davidiana_3        CGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [392]
P._tremula_1          CGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [392]
P._tremuloides_2      NGATCCATCTCGTTCATCAGGCAAAGCTGTTGTNGGATGGCTCAAGCTCA   [395]
P._davidiana_1        CGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [392]
P._tremuloides_1      TGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [393]
P._trichocarpa_1      TGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [395]
P._balsamifera_1      TGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [395]
P._balsamifera_2      TGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [393]
P._cathayana          TGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [384]
P._tristis            TGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [376]
P._angustifolia       TGATCCGTCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [392]
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P._trichocarpa_2      TGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [384]
P._balsamifera_3      TGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [385]
P._szechuanica        TGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [393]
P._nigra_3            TGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [392]
P._nigra_5            TGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [392]
P._nigra_1            TGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [390]
P._nigra_4            TGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [393]
P._nigra_2            TGATCCATCTCNTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [394]
S._integra            TGATCCATCTCCCTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [397]
S._nigra              TGATCCATCTCCCTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [394]
P._fremontii          TGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [393]
P._angulata           TGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [385]
P._laurifolia_1       TGATCCATCTCCTTCATCAGGCAAAGCCGTTGTCGGATGGCTCAAGCTCA   [396]
P._laurifolia_2       TGATCCATCTCCTTCATCAGGCAAAGCCGTTGTCGGATGGCTCAAGCTCA   [392]
P._simonii            TGATCCATCTCCTTCATCAGGCAAAGCCGTTGTCGGATGGCTCAAGCTCA   [393]
P._maximowiczii_1     TGATCCATCTCCTTCATCAGGCAAAGCCGTTGTCGGATGGCTCAAGCTCA   [395]
P._maximowiczii_2     TGATCCATCTCCTTCATCAGGCAAAGCCGTTGTCGGATGGCTCAAGCTCA   [395]
P._deltoides_1        TGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [161]
P._sargentii          TGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [393]
P._deltoides_2        TGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [395]
P._deltoides_3        TGATCCATCTCGTTCATCAGGCAAAGCTGTTGTCGGATGGCTCAAGCTCA   [395]

[                              410        420        430        440        450]
[                              .          .          .          .          .]

P._alba_1             AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [445]
P._alba_3             AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [445]
P._alba_2             AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [445]
P._grandidentata_2    AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [445]
P._grandidentata_3    AGGCAGCCCTGAGATGGGGNATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [445]
P._grandidentata_4    AGGCAGCCCTGAGATGGGGNATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [442]
P._davidiana_2        AGGCAGCCCTGAGATGGGGAATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [445]
P._davidiana_5        AGGCAGCCCTGAGATGGGGAATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [442]
P._davidiana_4        AGGCAGCCCTGAGATGGGGAATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [434]
P._davidiana_3        AGGCAGCCCTGAGATGGGGAATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [442]
P._tremula_1          AGGCAGCCCTGAGATGGGGAATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [442]
P._tremuloides_2      AGGCAGCCCTGAGATGGGGNATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [445]
P._davidiana_1        AGGCAGCCCTGAGATGGGGNATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [442]
P._tremuloides_1      AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [443]
P._trichocarpa_1      AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [445]
P._balsamifera_1      AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [445]
P._balsamifera_2      AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [443]
P._cathayana          AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [434]
P._tristis            AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [426]
P._angustifolia       AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [442]
P._trichocarpa_2      AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [434]
P._balsamifera_3      AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [435]
P._szechuanica        AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [443]
P._nigra_3            AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [442]
P._nigra_5            AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [442]
P._nigra_1            AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [440]
P._nigra_4            AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [443]
P._nigra_2            AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [444]
S._integra            AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCGGAGAGG   [447]
S._nigra              AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [444]
P._fremontii          AGGCAGCCCTGAGATGGGGCNTCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [443]
P._angulata           AGGCAGCCCTGAGATGGGGCGTCTTTGTCAGGAAGAAGGCTGCTGAGAGG   [435]
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P._laurifolia_1        AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG    [446]
P._laurifolia_2        AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG    [442]
P._simonii             AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG    [443]
P._maximowiczii_1      AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG    [445]
P._maximowiczii_2      AGGCAGCCCTGAGATGGGGCATCTTTGTCAGGAAGAAGGCTGCTGAGAGG    [445]
P._deltoides_1         AGGCAGCCCTGAGATGGGGNGTCTTTGTCAGGAAGAAGGCTGCTGAGAGG    [211]
P._sargentii           AGGCAGCCCTGAGATGGGGCGTCTTTGTCAGGAAGAAGGCTGCTGAGAGG    [443]
P._deltoides_2         AGGCAGCCCTGAGATGGGGCGTCTTTGTCAGGAAGAAGGCTGCTGAGAGG    [445]
P._deltoides_3         AGGCAGCCCTGAGATGGGGCGTCTTTGTCAGGAAGAAGGCTGCTGAGAGG    [445]

[                              460       470       480       490       500]
[                              .         .         .         .         .]

P._alba_1              CGAGCACAGCTCGTAGAGTTAGATGATTCTTAGATAGCCAATAGGGCAAT    [495]
P._alba_3              CGAGCACAGCTCGTAGAGTTAGATGATTCTTAGATAGCCAATAGGGCAAT    [495]
P._alba_2              CGAGCACAGCTCGTAGAGTTAGATGATTCTTAGATAGCCAATAGGGCAAT    [495]
P._grandidentata_2     CGAGCACAGCTCGTAGAGTTAGATGATTCTTAGATAGCCAATAGGGCAAT    [495]
P._grandidentata_3     CGAGCACAGCTCGTAGAGTTAGATGATTCTTAGATAGCCAATAGGGCAAT    [495]
P._grandidentata_4     CGAGCACAGCTCGTAGAGTTAGATGATTCTTAGATAGCCAATAGGGCAAT    [492]
P._davidiana_2         CGAGCACAGCTTGTAGAGTTAGATGATTCTTAGATAGCCAATAGGGCAAT    [495]
P._davidiana_5         CGAGCACAGCTTGTAGAGTTAGATGATTCTTAGATAGCCAATAGGGCAAT    [492]
P._davidiana_4         CGAGCACAGCTCGTAGAGTTAGATGATTCTTAGATAGCCAATAGGGCAAT    [484]
P._davidiana_3         CGAGCACAGCTTGTAGAGTTAGATGATTCTTAGATAGCCAATAGGGCAAT    [492]
P._tremula_1           CGAGCACAGCTTGTAGAGTTAGATGATTCTTAGATAGCCAATAGGGCAAT    [492]
P._tremuloides_2       CGAGCACAGCTCGTAGAGTTAGATGATTCTTAGATAGCCAATAGGGCAAT    [495]
P._davidiana_1         CGAGCACAGCTCGTAGAGTTAGATGATTCTTAGATAGCCAATAGGGCAAT    [492]
P._tremuloides_1       CGAGCACAGCTCGTAGAGTTAGATGATTCTTAGATAGCCAATAGGGCAAT    [493]
P._trichocarpa_1       CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [495]
P._balsamifera_1       CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [495]
P._balsamifera_2       CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [493]
P._cathayana           CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [484]
P._tristis             CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [476]
P._angustifolia        CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [492]
P._trichocarpa_2       CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [484]
P._balsamifera_3       CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [485]
P._szechuanica         CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [493]
P._nigra_3             CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [492]
P._nigra_5             CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [492]
P._nigra_1             CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [490]
P._nigra_4             CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [493]
P._nigra_2             CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [494]
S._integra             AGAGCACAGCTCGTTGAGTTGGATGATTCTTAGATAGCCCATATGGCAAT    [497]
S._nigra               CGAGCACAGCTCGTTGAGTTNGATGATTCTTAGATAGCCCATANGGCAAT    [494]
P._fremontii           CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [493]
P._angulata            CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [485]
P._laurifolia_1        CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [496]
P._laurifolia_2        CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [492]
P._simonii             CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [493]
P._maximowiczii_1      CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [495]
P._maximowiczii_2      CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [495]
P._deltoides_1         CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [261]
P._sargentii           CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [493]
P._deltoides_2         CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [495]
P._deltoides_3         CGAGCACAGCTCATAGAGTTAGATGATTCTTAGATAGGCAATAGGGCAAT    [495]

[                              510       520       530       540       550]
[                              .         .         .         .         .]
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P._alba_1               GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCAATCCCTAGC---T   [542]
P._alba_3               GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCAATCCCTAGC---T   [542]
P._alba_2               GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCAATCCCTAGC---T   [542]
P._grandidentata_2      GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCAATCCCTAGC---T   [542]
P._grandidentata_3      GTTGATCTGGTTTGGGTGNTAACAGGCTTCCTGCCCAATNCCTAGC---T   [542]
P._grandidentata_4      GTTGATCTGGTTTGGGTGNTAACAGGCTTCCTGCCCAATNCCTAGC---T   [539]
P._davidiana_2          GTTGATCTGGTTTGAGTGCTAACATGCTTCCTGCCCAATTCCTAGC---T   [542]
P._davidiana_5          GTTGATCTGGTTTGAGTGCTAACATGCTTCCTGCCCAATTCCTAGC---T   [539]
P._davidiana_4          GTTGATCTGGTTTGAGTGCTAACATGCTTCCTGCCCAATTCCTAGC---T   [531]
P._davidiana_3          GTTGATCTGGTTTGAGTGCTAACATGCTTCCTGCCCAATTCCTAGC---T   [539]
P._tremula_1            GTTGATCTGGTTTGAGTGCTAACATGCTTCCTGCCCAATTCCTAGC---T   [539]
P._tremuloides_2        GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCAATTCCTAGC---T   [542]
P._davidiana_1          GTTGATCTGGTTTGNGTGNTAACANGCTTCCTGCCCAATTCCTAGC---T   [539]
P._tremuloides_1        GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCAATTCCTAGC---T   [540]
P._trichocarpa_1        GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCAATTCCTGGC---T   [542]
P._balsamifera_1        GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCAATTCCTGGC---T   [542]
P._balsamifera_2        GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCAATTCCTGGC---T   [540]
P._cathayana            GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCAATTCCTGGC---T   [531]
P._tristis              GTTGATCTGGTTTGGGTGTTAAGAGGCTTCCTGCCCTATTCCTGGC---T   [523]
P._angustifolia         GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCAATTCCTGGC---T   [539]
P._trichocarpa_2        GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCAATTCCTGGC---T   [531]
P._balsamifera_3        GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCAATTCCTGGC---T   [532]
P._szechuanica          GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCAATTCCTGGC---T   [540]
P._nigra_3              GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCATTTCCTAGC---T   [539]
P._nigra_5              GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCATTTCCTAGC---T   [539]
P._nigra_1              GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCATTTCCTAGC---T   [537]
P._nigra_4              GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCATTTCCTAGC---T   [540]
P._nigra_2              GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCATTTCCTAGC---T   [541]
S._integra              GTTGATCTGGTCTGGGTGTTAACAGGCTTCCCCCCTAATTCCTAACTGAT   [547]
S._nigra                GTTGATCTGGTCTGGGTGTTAA----------------------------   [516]
P._fremontii            GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCANTTCCTAGC---T   [540]
P._angulata             GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCATTTCCTAGC---T   [532]
P._laurifolia_1         GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCATTTCCTAGC---T   [543]
P._laurifolia_2         GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCATTTCCTAGC---T   [539]
P._simonii              GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCATTTCCTAGC---T   [540]
P._maximowiczii_1       GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCATTTCCTAGC---T   [542]
P._maximowiczii_2       GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCATTTCCTAGC---T   [542]
P._deltoides_1          GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCAATTCCTAGC---T   [308]
P._sargentii            GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCAATTCCTAGC---T   [540]
P._deltoides_2          GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCAATTCCTAGC---T   [542]
P._deltoides_3          GTTGATCTGGTTTGGGTGTTAACAGGCTTCCTGCCCAATTCCTAGC---T   [542]

[                              560       570       580       590       600]
[                              .         .         .         .         .]

P._alba_1               CACTAGAGCACACACTGTTGTT----------------------------   [564]
P._alba_3               CACTAGAGCACACACTGTTGTTTTGCCTCATAATATTGGGGAGGCTCTTT   [592]
P._alba_2               CACTAGAGCACACACTGTTGTTTTGCCTCATAATATTGGGGAGGCTCTTT   [592]
P._grandidentata_2      CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGGAGGCTCTTT   [592]
P._grandidentata_3      CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCCGGCTCTTT   [592]
P._grandidentata_4      CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [589]
P._davidiana_2          CACTAGAGCAGACACTGTTGTTTTGCCTCGTAATATTGGGGAGGCTCTTT   [592]
P._davidiana_5          CACTAGAGCAGACACTGTTGTTTTGCCTCGTAATATTGGGGAGGCTCTTT   [589]
P._davidiana_4          CACTAGAGCAGACACTGTTGTTTTGCCTCGTAATATTGGGGAGGCTCTTT   [581]
P._davidiana_3          CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGGAGGCTCTTT   [589]
P._tremula_1            CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGGAGGCTCTTT   [589]
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P._tremuloides_2      CACTAGAGCAGACACTGTTGTTTTGCCTCATANTATTGGGGAGGCTCTTT   [592]
P._davidiana_1        CACNAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGNAGGCTCTTT   [589]
P._tremuloides_1      CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGGAGGCTCTTT   [590]
P._trichocarpa_1      CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [592]
P._balsamifera_1      CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [592]
P._balsamifera_2      CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [590]
P._cathayana          CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [581]
P._tristis            CACTAGAGCAGACGCTGTTGTTTTGCCTCATAATATTGGGGAGGCTCTTT   [573]
P._angustifolia       CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [589]
P._trichocarpa_2      CACTAGAGCCGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [581]
P._balsamifera_3      CACTAGAGCCGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [582]
P._szechuanica        CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [590]
P._nigra_3            CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [589]
P._nigra_5            CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [589]
P._nigra_1            CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [587]
P._nigra_4            CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [590]
P._nigra_2            CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [591]
S._integra            CACTCGAGCAGACGCTGTTGTTTTGCCTCGTAATATTGGGCAGGCTCTTC   [597]
S._nigra              ----------------------------------------------------  [516]
P._fremontii          CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [590]
P._angulata           CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [582]
P._laurifolia_1       CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [593]
P._laurifolia_2       CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [589]
P._simonii            CTCTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [590]
P._maximowiczii_1     CTCTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [592]
P._maximowiczii_2     CTCTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [592]
P._deltoides_1        CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [358]
P._sargentii          CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [590]
P._deltoides_2        CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [592]
P._deltoides_3        CACTAGAGCAGACACTGTTGTTTTGCCTCATAATATTGGGCAGGCTCTTT   [592]

[                              610       620       630       640       650]
[                              .         .         .         .         .]

P._alba_1             ----------------------------------------------------  [564]
P._alba_3             TGTTCAAGATTCTGCTGCTCTTAAACACCATTCTAGATGTCCAAGGATCT   [642]
P._alba_2             TGTTCAAGATTCTGCTGCTCTTAAACACCATTCTAGATGTCCAAGGATCT   [642]
P._grandidentata_2    TGTTCAAGATTCTGCTGCTCTTAGACACCATTCTAGATGTCCAAGGATCT   [642]
P._grandidentata_3    TGTTCAAGATTCTGCTGCTCTTAGACACCATTCTAGATGTCCAAGGATCT   [642]
P._grandidentata_4    TGTTCAAGATTCTGCTGCTCTTAGACACCATTCTAGATGTCCAAGGATCT   [639]
P._davidiana_2        TGTTCAAGATTCTGCTGCTCTTAGACACCATTCTAGATGTCCAAGGATCT   [642]
P._davidiana_5        TGTTCAAGATTCTGCTGCTCTTAGACACCATTCTAGATGTCCAAGGATCT   [639]
P._davidiana_4        TGTTCAAGATTCTGCTGCTCTTAGACACCATTCTAGATGTCCAAGGATCT   [631]
P._davidiana_3        TGTTCAAGATTCTGCTGCTCTTAGACACCATTCTAGATGTCCAAGGATCT   [639]
P._tremula_1          TGTTCAAGATTCTGCTGCTCTTAGACACCATTCTAGATGNCCAAGGATCT   [639]
P._tremuloides_2      TGTTCAAGATTCTGCTGCTCTTAGACACCATTCTAGATGTCCAAGGATCT   [642]
P._davidiana_1        TGTTCAAGATTCTGCTGCTCTTAGACACCATTCTAGATGTCCAAGGATCT   [639]
P._tremuloides_1      TGTTCAAGATTCTGCTGCTCTTAGACACCATTCTAGATGTCCAAGGATCT   [640]
P._trichocarpa_1      TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [642]
P._balsamifera_1      TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [642]
P._balsamifera_2      TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [640]
P._cathayana          TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [631]
P._tristis            TGTTCGAGGTTCTGCTGCTCTTAGACATCATTCAAGATGTCCGAGGATCT   [623]
P._angustifolia       TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [639]
P._trichocarpa_2      TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [631]
P._balsamifera_3      TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [632]
P._szechuanica        TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [640]
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P._nigra_3          TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [639]
P._nigra_5          TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [639]
P._nigra_1          TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [637]
P._nigra_4          TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [640]
P._nigra_2          TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [641]
S._integra          TGTTCAAGGTTCTGCTGCTCTTAGACATTAATCTAGATGTCCAAGGAT--   [645]
S._nigra            --------------------------------------------------   [516]
P._fremontii        TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGAT--   [638]
P._angulata         TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGAT--   [630]
P._laurifolia_1     TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [643]
P._laurifolia_2     TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [639]
P._simonii          TGTTCAAGGNTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [640]
P._maximowiczii_1   TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [642]
P._maximowiczii_2   TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [642]
P._deltoides_1      TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [408]
P._sargentii        TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [640]
P._deltoides_2      TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [642]
P._deltoides_3      TGTTCAAGGTTCTGCTGCTCTTAGACATCATTCTAGATGTCCAAGGATCT   [642]

[                           660       670       680       690       700]
[                           .         .         .         .         .]

P._alba_1           --------------------------------------------------   [564]
P._alba_3           T---AGTATTTCTACATTCCTCTGCTGTACAGTAAGCCAACTTCCTCCAT   [689]
P._alba_2           T---AGTATTTCTACATTCCTCTGCTGTACAGTAAGCCAACTTCCTCCAT   [689]
P._grandidentata_2  T---AGTATTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [689]
P._grandidentata_3  T---AGTATTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [689]
P._grandidentata_4  T---AGTATTTCTACATTCCTCTACTGTACAGTAAGCCNACTTCCTCCAT   [686]
P._davidiana_2      T---AGTATTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [689]
P._davidiana_5      T---AGTATTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [686]
P._davidiana_4      T---AGTATTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [678]
P._davidiana_3      T---AGTATTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [686]
P._tremula_1        T---AGTATTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [686]
P._tremuloides_2    T---AGTATTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [689]
P._davidiana_1      T---AGTATTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [686]
P._tremuloides_1    T---AGTATTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [687]
P._trichocarpa_1    T---AGTGTTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [689]
P._balsamifera_1    T---AGTGTTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [689]
P._balsamifera_2    T---AGTGTTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [687]
P._cathayana        T---AGTGTTTCTACATTCCTCTCCTGTACAGTAAGCCAACTTCCTCCAT   [678]
P._tristis          T---GTTGTTGCTTC----------------------------------   [635]
P._angustifolia     T---AGTGTTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [686]
P._trichocarpa_2    T---AGTGTTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [678]
P._balsamifera_3    T---AGTGTTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [679]
P._szechuanica      T---AGTGTTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [687]
P._nigra_3          T---AGTGTTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [686]
P._nigra_5          T---AGTGTTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [686]
P._nigra_1          T---AGTGTTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [684]
P._nigra_4          T---AGTGTTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [687]
P._nigra_2          T---AGTGTTTCTACATTCCTCTNCTGTACAGTAAGCCAACTTCCTCCAT   [688]
S._integra          ------------------------------------CAACTTCCTCCAT   [658]
S._nigra            --------------------------------------------------   [516]
P._fremontii        -------------------------------------------CTT-------   [641]
P._angulata         -----------------------------------------CT--------   [632]
P._laurifolia_1     T---AGTGTTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [690]
P._laurifolia_2     T---AGTGTTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [686]
P._simonii          T---AGTGTTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [687]
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P._maximowiczii_1     T---AGTGTTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [689]
P._maximowiczii_2     T---AGTGTTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [689]
P._deltoides_1        TCTTAGTGTTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [458]
P._sargentii          TCTTAGTGTTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [690]
P._deltoides_2        TCTTAGTGTTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [692]
P._deltoides_3        TCTTAGTGTTTCTACATTCCTCTACTGTACAGTAAGCCAACTTCCTCCAT   [692]

[                              710       720       730       740       750]
[                              .         .         .         .         .]

P._alba_1             ----------------------------------------------------   [564]
P._alba_3             CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [735]
P._alba_2             CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [735]
P._grandidentata_2    CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [735]
P._grandidentata_3    CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [735]
P._grandidentata_4    CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [732]
P._davidiana_2        CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [735]
P._davidiana_5        CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [732]
P._davidiana_4        CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [724]
P._davidiana_3        CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [732]
P._tremula_1          CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [732]
P._tremuloides_2      CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCCTGGTGAAGTTGCT   [735]
P._davidiana_1        CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCNTGGTGAAGTTGCT   [732]
P._tremuloides_1      CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCCTGGTGAAGTTGCT   [733]
P._trichocarpa_1      CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [735]
P._balsamifera_1      CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [735]
P._balsamifera_2      CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [733]
P._cathayana          CAACAGCTGTCCTGCAGTTGCCCCTTCT----------------------   [706]
P._tristis            ----------------------------------------------------   [635]
P._angustifolia       CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [732]
P._trichocarpa_2      CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [724]
P._balsamifera_3      CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [725]
P._szechuanica        CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [733]
P._nigra_3            CAACAGCTGTCCTGCAGTTGCCCCTTCTCGATCCCCTTGGTGAAGTTGCT   [736]
P._nigra_5            CAACAGCTGTCCTGCAGTTGCCCCTTCTCGATCCCCTTGGTGAAGTTGCT   [736]
P._nigra_1            CAACAGCTGTCCTGCAGTTGCCCCTTCTCGATCCCCTTGGTGAAGTTGCT   [734]
P._nigra_4            CAACAGCTGTCCTGCAGTTGCCCCTTCTCGATCCCCTTGGTGAAGTTGCT   [737]
P._nigra_2            CAACAGCTGTCCTGCAGTTGCCCCTTCTCGATCCCCTTGGTGAAGTTGCT   [738]
S._integra            CAACAGCTGTTCTGCAGTTG--TCTTCTC---CCCCTTTT-GAAGTTGCT   [702]
S._nigra              ----------------------------------------------------   [516]
P._fremontii          ----------------------------------------------------   [641]
P._angulata           ----------------------------------------------------   [632]
P._laurifolia_1       CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [736]
P._laurifolia_2       CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [732]
P._simonii            CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [733]
P._maximowiczii_1     CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [735]
P._maximowiczii_2     CAACAGCTGTCCTGCAGTTGCCCCTTCT----CCCCTTGGTGAAGTTGCT   [735]
P._deltoides_1        CAACAGCTG-CCTGCAGTTGCCCCATCT----CCCCTTGGTGAAGTTGCT   [503]
P._sargentii          CAACAGCTG-CCTGCAGTTGCCCCATCT----CCCCTTGGTGAAGTTGCT   [735]
P._deltoides_2        CAACAGCTG-CCTGCAGTTGCCCCATCT----CCCCTTGGTGAAGTTGCT   [737]
P._deltoides_3        CAACAGCTG-CCTGCAGTTGCCCCATCT----CCCCTTGGTGAAGTTGCT   [737]

[                              760       770       780       790       800]
[                              .         .         .         .         .]

P._alba_1             ----------------------------------------------------   [564]
P._alba_3             TACCTTCCAGT-CCAGCACTCT-AACAGTAGTGAATTCTGAAGATTGACG   [783]
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P._alba_2               TACCTTCCAGT-CCAGCACTCT-AACAGTAGTGAATTCTGAAGATTGACG   [783]
P._grandidentata_2      TACCTTCCAGT-CCAGCACTCT-AACAGTAGTGAATTCTGAAGATTGACG   [783]
P._grandidentata_3      TACCTTCCAGT-CCAGCACTCT-AACAGTAGTGAATTCTGAAGATTGACG   [783]
P._grandidentata_4      TACCTTCCAGT-CCAGCACTCT-AACAGTAGTGAATTCTGAAGATTGACG   [780]
P._davidiana_2          TACCTTCCAGT-CCAGCACTCTTAACAGTAGTGAATTCTGAAGATTGACG   [784]
P._davidiana_5          TACCTTCCAGT-CCAGCACTCTTAACAGTAGTGAATTCTGAAGATTGACG   [781]
P._davidiana_4          TACCTTCCAGT-CCAGCACTC----------------------------   [744]
P._davidiana_3          TACCTTCCAGT-CCAGCACTC----------------------------   [752]
P._tremula_1            TACCTTCCAGT-CCAGCACTCT-AACAGTAGTGAATTCTGAAGATTGACG   [780]
P._tremuloides_2        TACCTTCCAGT-CCAGCACTCT-AACAGTAGTGAATTCTGAAGATTGACG   [783]
P._davidiana_1          TACCTTCCAGT-CCAGCACTCT-AACAGTAGTGAATTCTGAAGATTGACG   [780]
P._tremuloides_1        TACCTTCCAGT-CCAGCACTCT-AACAGTAGTGAATTCTGAAGATTGACG   [781]
P._trichocarpa_1        TACCTTCCAGT-CCAGCGCTCT-AACAGTAGTGAATTCTGAAGGTTGACG   [783]
P._balsamifera_1        TACCTTCCAGT-CCAGCGCTCT-AACAGTAGTGAATTCTGAAGGTTGACG   [783]
P._balsamifera_2        TACCTTCCAGT-CCAGCGCTCT-AACAGTAGTGAATTCTGAAGGTTGACG   [781]
P._cathayana            ---------------------------------------------------   [706]
P._tristis              ---------------------------------------------------   [635]
P._angustifolia         TACCTTCCAGT-CCAGCGCTCT-AACAGTAGTGAATTCTGAAGGTTGACG   [780]
P._trichocarpa_2        TACCTTCCAGT-CCAGCGCTCT-AACAGTAGTGAATTCTGAAGGTTGACG   [772]
P._balsamifera_3        TACCTTCCAGT-CCAGCGCTCT-AACAGTAGTGAATTCTGAAGGTTGACG   [773]
P._szechuanica          TACCTTCCAGT-CCAGCGCTCT-AACAGTAGTGAATTCTGAAGGTTGACG   [781]
P._nigra_3              TACCTTCCAGT-CCAGCACTCT-AACGGTAGTGAATTCTGAAGGTTGACG   [784]
P._nigra_5              TACCTTCCAGT-CCAGCACTCT-AACGGTAGTGAATTCTGAAGGTTGACG   [784]
P._nigra_1              TACCTTCCAGT-CCAGCACTCT-AACGGTAGTGAATTCTGAAGGTTGACG   [782]
P._nigra_4              TACCTTCCAGT-CCAGCACTCT-AACGGTAGTGAATTCTGAAGGTTGACG   [785]
P._nigra_2              TACCTTCCAGT-CCAGCACTCT-AACGGTAGTGAATTCTGAAGGTTGACG   [786]
S._integra              TCTCTTCCAGTTCCAGCACTCT-AACGGTAGTGAATTCTGACGGNTGACT   [751]
S._nigra                ---------------------------------------------------   [516]
P._fremontii            ---------------------------------------------------   [641]
P._angulata             ---------------------------------------------------   [632]
P._laurifolia_1         TACCTTCCAGT-CCAGCACTCT-AACAGTAATGAATTCTGAAGGTTGACG   [784]
P._laurifolia_2         TACCTTCCAGT-CCAGCACTCT-AACAGTAATGAATTCTGAAGGTTGACG   [780]
P._simonii              TACCTTCCAGT-CCAGCACTCT-AACAGTAATGAATTCTGAAGGTTGACG   [781]
P._maximowiczii_1       TACCTTCCAGT-CCAGCACTCT-AACAGTAATGAATTCTGAAGGTTGACG   [783]
P._maximowiczii_2       TACCTTCCAGT-CCAGCACTCT-AACAGTAATGAATTCTGAAGGTTGACG   [783]
P._deltoides_1          TACCTTCCAGT-CCAGCACTCT-AACAGTAGTGAAATCTGAAGGTTGACG   [551]
P._sargentii            TACCTTCCAGT-CCAGCACTCT-AACAGTAGTGAAATCTGAAGGTTGACG   [783]
P._deltoides_2          TACCTTCCAGT-CCAGCACTCT-AACAGTAGTGAAATCTGAAGGTTGACG   [785]
P._deltoides_3          TACCTTCCAGT-CCAGCACTCT-AACAGTAGTGAAATCTGAAGGTTGACG   [785]

[                              810       820       830       840       850]
[                              .         .         .         .         .]

P._alba_1               ---------------------------------------------------   [564]
P._alba_3               GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [833]
P._alba_2               GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [833]
P._grandidentata_2      GAGTTGAGCTGTTTCCATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [833]
P._grandidentata_3      GAGTTGAGCTGTTTCNATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [833]
P._grandidentata_4      GAGTTGAGCTGTTTCNATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [830]
P._davidiana_2          GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [834]
P._davidiana_5          GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [831]
P._davidiana_4          ---------------------------------------------------   [744]
P._davidiana_3          ---------------------------------------------------   [752]
P._tremula_1            GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [830]
P._tremuloides_2        GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGNTGGAGACTTCTAA   [833]
P._davidiana_1          GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [830]
P._tremuloides_1        GAGTTGAGCTGTTTCTATNGANGATATTTTTGGATGGTGGAGACTTCTAA   [831]
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P._trichocarpa_1      GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [833]
P._balsamifera_1      GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [833]
P._balsamifera_2      GAGTTGAGCTGTTTCTATTGACGATATTTTTGG----------------   [814]
P._cathayana          --------------------------------------------------   [706]
P._tristis            --------------------------------------------------   [635]
P._angustifolia       GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [830]
P._trichocarpa_2      GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [822]
P._balsamifera_3      GAGTTGAGCTGTTTCTATTGACGATATTTTTGGAT---------------   [808]
P._szechuanica        GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCNAA   [831]
P._nigra_3            GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [834]
P._nigra_5            GAATTGAGCTGTTTCTATTGACGATATTTTTGGA----------------   [818]
P._nigra_1            GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [832]
P._nigra_4            GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [835]
P._nigra_2            GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [836]
S._integra            GAGTTGAGCTGTTTCTGCTGACCAT------------------------   [776]
S._nigra              --------------------------------------------------   [516]
P._fremontii          --------------------------------------------------   [641]
P._angulata           --------------------------------------------------   [632]
P._laurifolia_1       GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [834]
P._laurifolia_2       GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [830]
P._simonii            GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [831]
P._maximowiczii_1     GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [833]
P._maximowiczii_2     GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [833]
P._deltoides_1        GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [601]
P._sargentii          GAGTTGAGCTGTTTCTATTGACNATATTTTTGGATGGTGGAGACTTCTAA   [833]
P._deltoides_2        GAGTTGAGCTGTTTCTATTGACNATATTTTTGGATGGTGGAGACTTCTAA   [835]
P._deltoides_3        GAGTTGAGCTGTTTCTATTGACGATATTTTTGGATGGTGGAGACTTCTAA   [835]

[                              860       870       880       890       900]
[                              .         .         .         .         .]

P._alba_1             --------------------------------------------------   [564]
P._alba_3             TATTTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [883]
P._alba_2             TATTTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [883]
P._grandidentata_2    TATTTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [883]
P._grandidentata_3    TATTTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [883]
P._grandidentata_4    TATTTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [880]
P._davidiana_2        TATTTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [884]
P._davidiana_5        TATTTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [881]
P._davidiana_4        --------------------------------------------------   [744]
P._davidiana_3        --------------------------------------------------   [752]
P._tremula_1          TATTTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [880]
P._tremuloides_2      TATTTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [883]
P._davidiana_1        TATTTCGGNCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [880]
P._tremuloides_1      TATTTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [881]
P._trichocarpa_1      TATTTCGGCCATCCATTCCCGAAGCT------------------------   [859]
P._balsamifera_1      TATTTCGGCCATCCATTCCCGAAGCTG-----------------------   [860]
P._balsamifera_2      --------------------------------------------------   [814]
P._cathayana          --------------------------------------------------   [706]
P._tristis            --------------------------------------------------   [635]
P._angustifolia       TATTTCGGCCATCCATTCCCG------------------------------   [851]
P._trichocarpa_2      TATTTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [872]
P._balsamifera_3      --------------------------------------------------   [808]
P._szechuanica        TATTTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [881]
P._nigra_3            TATTTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [884]
P._nigra_5            --------------------------------------------------   [818]
P._nigra_1            TATTTCGGCC----------------------------------------   [842]
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P._nigra_4              TATTTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [885]
P._nigra_2              TATTTCGGNCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [886]
S._integra              ----------CCATTCCCGAAGCTGGA---CTCAGCACGGGCGCGTCT   [811]
S._nigra                ----------------------------------------------   [516]
P._fremontii            ----------------------------------------------   [641]
P._angulata             ----------------------------------------------   [632]
P._laurifolia_1         TATTTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [884]
P._laurifolia_2         TATTTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [880]
P._simonii              TATTTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [881]
P._maximowiczii_1       TATTTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [883]
P._maximowiczii_2       TATTTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCACGGGTGCATCT   [883]
P._deltoides_1          TATCTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCGCGGGTGCATCT   [651]
P._sargentii            TATCTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCGCGGGTGCATCT   [883]
P._deltoides_2          TATCTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCGCGGGTGCATCT   [885]
P._deltoides_3          TATCTCGGCCATCCATTCCCGAAGCTGGAAAACTCAGCGCGGGTGCATCT   [885]

[                                910       920       930       940       950]
[                                .         .         .         .         .]

P._alba_1               ----------------------------------------------   [564]
P._alba_3               CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTAATTGCATCA   [933]
P._alba_2               CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTAATTGCATCA   [933]
P._grandidentata_2      CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTAATTGCATCA   [933]
P._grandidentata_3      CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTAATTGCATCA   [933]
P._grandidentata_4      CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTAATTGCATCA   [930]
P._davidiana_2          CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTCATTGCATCA   [934]
P._davidiana_5          CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTCATTGCATCA   [931]
P._davidiana_4          ----------------------------------------------   [744]
P._davidiana_3          ----------------------------------------------   [752]
P._tremula_1            CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTNATTGCATCA   [930]
P._tremuloides_2        CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTNATTGCATCA   [933]
P._davidiana_1          CNGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTAATTGCATCA   [930]
P._tremuloides_1        CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTAATTGCATCA   [931]
P._trichocarpa_1        ----------------------------------------------   [859]
P._balsamifera_1        ----------------------------------------------   [860]
P._balsamifera_2        ----------------------------------------------   [814]
P._cathayana            ----------------------------------------------   [706]
P._tristis              ----------------------------------------------   [635]
P._angustifolia         ----------------------------------------------   [851]
P._trichocarpa_2        C---------------------------------------------   [873]
P._balsamifera_3        ----------------------------------------------   [808]
P._szechuanica          CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCATCCTAATTGCATCA   [931]
P._nigra_3              CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTAATTGCATCA   [934]
P._nigra_5              ----------------------------------------------   [818]
P._nigra_1              ----------------------------------------------   [842]
P._nigra_4              CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTAATTGCATCA   [935]
P._nigra_2              CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTAATTGCATCA   [936]
S._integra              CGGTTTCTGCCAGGAAGTAGATAAAATAGTAATGT--CTTAATTGCATCA   [859]
S._nigra                ----------------------------------------------   [516]
P._fremontii            ----------------------------------------------   [641]
P._angulata             ----------------------------------------------   [632]
P._laurifolia_1         CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTAATTGCATCA   [934]
P._laurifolia_2         CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTAATTGCATCA   [930]
P._simonii              CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTAATTGCATCT   [931]
P._maximowiczii_1       CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTAATTGCATCT   [933]
P._maximowiczii_2       CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTAATTGCATCT   [933]
P._deltoides_1          CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTAATTGCATCA   [701]
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P._sargentii          CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTAATTGCATCA   [933]
P._deltoides_2        CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTAATTGCATCA   [935]
P._deltoides_3        CGGTTTCTGCCAGGAAGTAGATAGAATAGAAATGCAGCCTAATTGCATCA   [935]

[                              960       970       980       990       1000]
[                              .         .         .         .         .]

P._alba_1             ----------------------------------------------------   [564]
P._alba_3             CCTCCGTATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTGTACAG   [983]
P._alba_2             CCTCCGTATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTGTACAG   [983]
P._grandidentata_2    CCTCCATATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTTTACAG   [983]
P._grandidentata_3    CCTCCNTATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTTTACAG   [983]
P._grandidentata_4    CCTCCNTATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTTTACAG   [980]
P._davidiana_2        CCTCCGTATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTGTACAG   [984]
P._davidiana_5        CCTCCGTATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTGTACAG   [981]
P._davidiana_4        ----------------------------------------------------   [744]
P._davidiana_3        ----------------------------------------------------   [752]
P._tremula_1          CCTCCGTATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTGTACAG   [980]
P._tremuloides_2      CCTCCGTATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTCTGTACAG   [983]
P._davidiana_1        CCTCCGTATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTGTACAG   [980]
P._tremuloides_1      CCTCCGTATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTCTGTACAG   [981]
P._trichocarpa_1      ----------------------------------------------------   [859]
P._balsamifera_1      ----------------------------------------------------   [860]
P._balsamifera_2      ----------------------------------------------------   [814]
P._cathayana          ----------------------------------------------------   [706]
P._tristis            ----------------------------------------------------   [635]
P._angustifolia       ----------------------------------------------------   [851]
P._trichocarpa_2      ----------------------------------------------------   [873]
P._balsamifera_3      ----------------------------------------------------   [808]
P._szechuanica        CCTCCGTATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTGTACAG   [981]
P._nigra_3            CCTCCGTATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTGTACAT   [984]
P._nigra_5            ----------------------------------------------------   [818]
P._nigra_1            ----------------------------------------------------   [842]
P._nigra_4            CCTCCGTATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTGTACAT   [985]
P._nigra_2            CCTCCGTATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTGTACAT   [986]
S._integra            CCTTCGTGCATGGCTAGTAGGTCCTGTCTTAGTGTGCTTTGTTTGTACAT   [909]
S._nigra              ----------------------------------------------------   [516]
P._fremontii          ----------------------------------------------------   [641]
P._angulata           ----------------------------------------------------   [632]
P._laurifolia_1       CCTCCGTATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTGTACAT   [984]
P._laurifolia_2       CCTCCGTATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTGTACAT   [980]
P._simonii            CCTCCGTATATGGCTAGTGGGTCCTATCTTCGTGTGTTTTNTTTGTACAG   [981]
P._maximowiczii_1     CCTCCGTATATGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTGTACAG   [983]
P._maximowiczii_2     CCTCCGTATATGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTGTACAG   [983]
P._deltoides_1        CCTCCGTATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTGTACAG   [751]
P._sargentii          CCTCCGTATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTGTACAG   [983]
P._deltoides_2        CCTCCGTATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTGTACAG   [985]
P._deltoides_3        CCTCCGTATTTGGCTAGTGGGTCCTATCTTCGTGTGTTTTGTTTGTACAG   [985]

[                              1010      1020      1030      1040      1050]
[                              .         .         .         .         .]

P._alba_1             ----------------------------------------------------   [564]
P._alba_3             GGTCAGCAAAAACGAACGGCCAGCTT-----TTGTTGTTCCCAAGTAACA   [1028]
P._alba_2             GGTCAGCAAAAACGAACGGCCAGCTT-----TTGTTGTTCCCAAGTAACA   [1028]
P._grandidentata_2    GGTCAACAAAAACGAACGGCCAACTT-----TTGTTGTTCCCTAGTACCA   [1028]
P._grandidentata_3    GGTCAACAAAAACGAACGGCCAACTT-----TTGTTGTTCCCTAGTANCA   [1028]
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P._grandidentata_4    GGTCAACAAAAACGAACGGCCAACTT-----TTGTTGTTCCCTAGTANCA   [1025]
P._davidiana_2        GGTCAACAAAAACGAACGGCCAACTT-----TTGTTGTTCCCTAGTAACA   [1029]
P._davidiana_5        GGTCAACAAAAACGAACGGCCAACTT-----TTGTTGTTCCCTAGTAACA   [1026]
P._davidiana_4        ----------------------------------------------------  [744]
P._davidiana_3        ----------------------------------------------------  [752]
P._tremula_1          GGGCAACAAAAACGAACGGCCAACTT-----TTGTTGTTCCCTAGTAACA   [1025]
P._tremuloides_2      GGTCAACAAAAACGAACGGCCAACTT-----TTGTTGTTCCCTAGTAACA   [1028]
P._davidiana_1        GGTCAACAAAAACGAACGGCCAACTT-----TTGTTGTTCCCTAGTAACA   [1025]
P._tremuloides_1      GGTCAACAAAAACGAACGGCCAACTT-----TTGTTGTTCCCTAGTAACA   [1026]
P._trichocarpa_1      ----------------------------------------------------  [859]
P._balsamifera_1      ----------------------------------------------------  [860]
P._balsamifera_2      ----------------------------------------------------  [814]
P._cathayana          ----------------------------------------------------  [706]
P._tristis            ----------------------------------------------------  [635]
P._angustifolia       ----------------------------------------------------  [851]
P._trichocarpa_2      ----------------------------------------------------  [873]
P._balsamifera_3      ----------------------------------------------------  [808]
P._szechuanica        GGTCAACAAAAACGANCGGCCAACTT-----TTGTTGTTCCTTAGTAACA   [1026]
P._nigra_3            GGTCAACAAAAACGAACGGCCAACTT-----TTGTTGTTCCTTAGTAACA   [1029]
P._nigra_5            ----------------------------------------------------  [818]
P._nigra_1            ----------------------------------------------------  [842]
P._nigra_4            GGTCAACAAAAACGAACGGCCAACTT-----TTGTTGTTCCTTAGTAACA   [1030]
P._nigra_2            GGTCAACAAAAACGAACGGCCAACTT-----TTGTTGTTCCTTAGTAACA   [1031]
S._integra            GGTCAACAAAAACGAGCGNCCAACTTAACTTTTGTTGTTCCTTAGTAACA   [959]
S._nigra              ----------------------------------------------------  [516]
P._fremontii          ----------------------------------------------------  [641]
P._angulata           ----------------------------------------------------  [632]
P._laurifolia_1       GGTCAACAAAAACGAACGGCCAACTT-----TTGTTGTTCCTTAGTAACA   [1029]
P._laurifolia_2       GGTCAACAAAAACGAACGGCCAACTT-----TTGTTGTTCCTTAGTAACA   [1025]
P._simonii            GGTCAACAAAAACGAACGGCCAACTT-----TTGTTGTTCCTTAGTAACA   [1026]
P._maximowiczii_1     GGTCAACAAAAACGAACGGCCAACTT-----TTGTTGTTCCTTAGTAACA   [1028]
P._maximowiczii_2     GGTCAACAAAAACGAACGGCCAACTT-----TTGTTGTTCCTTAGTAACA   [1028]
P._deltoides_1        GGTCAACAAAAACGAACGGCCAACTT-----TTGGTGTTCCTTAGTAACA   [796]
P._sargentii          GGTCAACAAAAACGAACGGCCAACTT-----TTGGTGTTCCTTAGTAACA   [1028]
P._deltoides_2        GGTCAACAAAAACGAACGGCCAACTT-----TTGGTGTTCCTTAGTAACA   [1030]
P._deltoides_3        GGTCAACAAAAACGAACGGCCAACTT-----TTGGTGTTCCTTAGTAACA   [1030]

[                              1060      1070      1080      1090      1100]
[                              .         .         .         .         .]

P._alba_1             ----------------------------------------------------  [564]
P._alba_3             TTAGAATAATTGTTGGTACCAGTGATGAAGTTGGCCGGAACAAAAA-CCT   [1077]
P._alba_2             TTAGAATAATTGTTGGTACCAGTGATGAAGTTGGCCGGAACAAAAA-CCT   [1077]
P._grandidentata_2    TTAGAATAATTGTTGGTACCAGTGATGAAGTTGGCCGGAACAAAAA-CCT   [1077]
P._grandidentata_3    TTAGAATAATTGTTGGTACCAGTGATGAAGTTGGCCGGAACAAAAA-CCT   [1077]
P._grandidentata_4    TTAGAATAATTGTTGGTACCAGTGATGAAGTTGGCCGGAACAAAAA-CCT   [1074]
P._davidiana_2        TTAGAATAATTGTTGGTACCAGTGATGAAGTTGGCCGGAACAAAAA-CCT   [1078]
P._davidiana_5        TTAGAATAATTGTTGGTACCAGTGATGAAGTTGGCCGGAACAAAAA-CCT   [1075]
P._davidiana_4        ----------------------------------------------------  [744]
P._davidiana_3        ----------------------------------------------------  [752]
P._tremula_1          TTAGAATAATTGTTGGTACCAGTGATNAAGTTGGCCGGAACAAAAA-CCT   [1074]
P._tremuloides_2      TTAGAATAATTGTTGGTACCAGTGATGAAGTTGGCCGGAACAAAAA-CCT   [1077]
P._davidiana_1        TTAGAATAATTGTTGGTACCAGTGATGAAGTTGGCCGGAACAAAAA-CCT   [1074]
P._tremuloides_1      TTAGAATAATTGTTGGTACCAGTGATGAAGTTGGCCGGAACAAAAA-CCT   [1075]
P._trichocarpa_1      ----------------------------------------------------  [859]
P._balsamifera_1      ----------------------------------------------------  [860]
P._balsamifera_2      ----------------------------------------------------  [814]
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P._cathayana            ------------------------------------------------  [706]
P._tristis              ------------------------------------------------  [635]
P._angustifolia         ------------------------------------------------  [851]
P._trichocarpa_2        ------------------------------------------------  [873]
P._balsamifera_3        ------------------------------------------------  [808]
P._szechuanica          TTAGAATAATTGCTGGTACCAGTGATGAAGTTGGCCGGAACAAAAGACCT  [1076]
P._nigra_3              TTAGAATAATTGCTGGTACCAGTGATGAATTTGGCCGGAACAAAAA-CCT  [1078]
P._nigra_5              ------------------------------------------------  [818]
P._nigra_1              ------------------------------------------------  [842]
P._nigra_4              TTAGAATAATTGCTGGTACCAGTGATGAATTTGGCCGGAACAAAAA-CCT  [1079]
P._nigra_2              TTAGAATAATTGCTGGTACCAGTGATGAATTTGGCCGGAACAAAAA-CCT  [1080]
S._integra              TTAAAATAAATGTTAGCACCAGCGAAGAAGTTGGCCAGATAAAAAAAC--  [1007]
S._nigra                ------------------------------------------------  [516]
P._fremontii            ------------------------------------------------  [641]
P._angulata             ------------------------------------------------  [632]
P._laurifolia_1         TTAGAATAATTGCTGGTACCAGTGATGAAGTTGGCCGGAACAAAAA-CCT  [1078]
P._laurifolia_2         TTAGAATAATTGCTGGTACCAGTGATGAAGTTGGCCGGAACAAAAA-CCT  [1074]
P._simonii              TTAGAATAATTGCTGGTACCAGTGATGAAGTTGGCCGGAACAAAAA-CNT  [1075]
P._maximowiczii_1       TTAGAATAATTGCTGGTACCAGTGATGAAGTTGGCCGGAACAAAAA-CCT  [1077]
P._maximowiczii_2       TTAGAATAATTGCTGGTACCAGTGATGAAGTTGGCCGGAACAAAAA-CCT  [1077]
P._deltoides_1          TTAGAATAATTGCTGGTACCAGTGATGAAGTTGGCCGGAACAAAAA-CCT  [845]
P._sargentii            TTAGAATAATTGCTGGTACCAGTGATGAAGTTGGCCGGAACAAAAA-CCT  [1077]
P._deltoides_2          TTAGAATAATTGCTGGTACCAGTGATGAAGTTGGCCGGAACAAAAA-CCT  [1079]
P._deltoides_3          TTAGAATAATTGCTGGTACCAGTGATGAAGTTGGCCGGAACAAAAA-CCT  [1079]

[                              1110      1120      1130      1140      1150]
[                              .         .         .         .         .]

P._alba_1               ------------------------------------------------  [564]
P._alba_3               CAATTCAGGCAGAAACTCTTACGTTGTTTGTGAATTGATGAAGAGTCTTC  [1127]
P._alba_2               CAATTCAGGCAGAAACTCTTACGTTGTTTGTGAATTGATGAAGAGTCTTC  [1127]
P._grandidentata_2      CAATTCAGGCAGAAACTCTTACGTTGTTTGTGAATTGATGAAGAGTCTTC  [1127]
P._grandidentata_3      CAATTCAGGCAGAAACTCTTACGTTGTTTGTGAATTGATGAAGAGTCTTC  [1127]
P._grandidentata_4      CAATTCAGGCAGAAACTCTTACGTTGTTTGTGAATTGATGAAGAGTCTTC  [1124]
P._davidiana_2          CAATTCAGGCAGAAACTCTTACGTTGTTTGTGAATTGATGAAGAGTCTTC  [1128]
P._davidiana_5          CAATTCAGGCAGAAACTCTTACGTTGTTTGTGAATTGATGAAGAGTCTTC  [1125]
P._davidiana_4          ------------------------------------------------  [744]
P._davidiana_3          ------------------------------------------------  [752]
P._tremula_1            CAATTCAGGCAGAAACTCTTACGTTGTTTGTGAATTGATGAAGAGTCTTC  [1124]
P._tremuloides_2        CAATTCAGGCAGAAACTCTTACGTTGTTTGTGAATTGATGAAGAGTCTTC  [1127]
P._davidiana_1          CAATTCAGGCAGAAACTCTTACGTTGTTTGTGAATTGATGAAGAGTCTTC  [1124]
P._tremuloides_1        CAATTCAGGCNGAAACTCTTACGTTGTTTGTGAATTGATGAAGAGTCTNC  [1125]
P._trichocarpa_1        ------------------------------------------------  [859]
P._balsamifera_1        ------------------------------------------------  [860]
P._balsamifera_2        ------------------------------------------------  [814]
P._cathayana            ------------------------------------------------  [706]
P._tristis              ------------------------------------------------  [635]
P._angustifolia         ------------------------------------------------  [851]
P._trichocarpa_2        ------------------------------------------------  [873]
P._balsamifera_3        ------------------------------------------------  [808]
P._szechuanica          CAATTCAGGCAGAAACTCTTACGTTGTTTGTGAATTGATGAAGAGTCTTC  [1126]
P._nigra_3              CAATTCAGGCAGAAACTCTTACGTTGTTTGTGAATTGATGAAGAGTCTTC  [1128]
P._nigra_5              ------------------------------------------------  [818]
P._nigra_1              ------------------------------------------------  [842]
P._nigra_4              CAATTCAGGCAGAAACTCTTACGTTGTTTGTGAATTGATGAAGAGTCTTC  [1129]
P._nigra_2              CAATTCAGGCAGAAACTCTTACGTTGTTTGTGAATTGATGAAGAGTCTTC  [1130]
S._integra              ------------------------------------------------  [1007]
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```
S._nigra                ------------------------------------------------   [516]
P._fremontii            ------------------------------------------------   [641]
P._angulata             ------------------------------------------------   [632]
P._laurifolia_1         CAATTCAGGCAGAAACTCTTACATTGTTTGTGAATTGATGAAGAGTCTTC   [1128]
P._laurifolia_2         CAATTCAGGCAGAAACTCTTACATTGTTTGTGAATTGATGAAGAGTCTTC   [1124]
P._simonii              CAATTCAGGCAGAAACTCGTACGTTGTTTGTGAATTGATGAAGAGTCTTC   [1125]
P._maximowiczii_1       CAATTCAGGCAGAAACTCTTACGTTGTTTGTGAATTGATGAAGAGTCTTC   [1127]
P._maximowiczii_2       CAATTCAGGCAGAAACTCTTACGTTGTTTGTGAATTGATGAAGAGTCTTC   [1127]
P._deltoides_1          CAATTCAGGCAGAAACTCTTACGTTGTCTGTGAATTGATGAAGAGTCTTC   [895]
P._sargentii            CAATTCAGGCAGAAACTCTTACGTTGTCTGTGAATTGATGAAGAGTCTTC   [1127]
P._deltoides_2          CAATTCAGGCAGAAACTCTTACGTTGTCTGTGAATTGATGAAGAGTCTTC   [1129]
P._deltoides_3          CAATTCAGGCAGAAACTCTTACGTTGTCTGTGAATTGATGAAGAGTCTTC   [1129]

[                                1160      1170      1180      1190      1200]
[                                .         .         .         .         .]

P._alba_1               ------------------------------------------------   [564]
P._alba_3               ACGTAATGAAATAAGATTGATTTATGGCTGGAT-ATCAT-----------   [1165]
P._alba_2               ACGTAATGAAATAAGATTGATTTATGGCTGGAT-ATCAT-----------   [1165]
P._grandidentata_2      ACGTAATGAAATAAGATTGATTTATGGCTGGAT-----------------   [1160]
P._grandidentata_3      ACGTAATGAAATAAGATTGATTTATGGCTGG-------------------   [1158]
P._grandidentata_4      ACGTAATGAAATAAGATTGATTTATGGCTGGA------------------   [1156]
P._davidiana_2          ACGTAATGAAATAAGATTGATTTATGGCTGGAT-ATCAT-----------   [1166]
P._davidiana_5          ACGTAATGAAATAAGATTGATTTATGG-----------------------   [1152]
P._davidiana_4          ------------------------------------------------   [744]
P._davidiana_3          ------------------------------------------------   [752]
P._tremula_1            ACGTAATGAAATAAGATTGATTTATGGCTGGA------------------   [1156]
P._tremuloides_2        ACGTAATGAAATAAGATTGATTTATGGCTG--------------------   [1157]
P._davidiana_1          ACGTAATGAAATAAGATTGATTCATGGCTGGAT-----------------   [1157]
P._tremuloides_1        ACGTAATGAAATAAGATTG-------------------------------   [1144]
P._trichocarpa_1        ------------------------------------------------   [859]
P._balsamifera_1        ------------------------------------------------   [860]
P._balsamifera_2        ------------------------------------------------   [814]
P._cathayana            ------------------------------------------------   [706]
P._tristis              ------------------------------------------------   [635]
P._angustifolia         ------------------------------------------------   [851]
P._trichocarpa_2        ------------------------------------------------   [873]
P._balsamifera_3        ------------------------------------------------   [808]
P._szechuanica          ACGTAATGAAATAAGATTGATTTATGGCTGGA------------------   [1158]
P._nigra_3              ACGTAATGAAATAAGATTGATTTATGGCTGGATTATCATTGGCTAACATA   [1178]
P._nigra_5              ------------------------------------------------   [818]
P._nigra_1              ------------------------------------------------   [842]
P._nigra_4              ACGTAATGAAATAAGATTGATTTATGGCTGGATTATCATTGGCTAACATA   [1179]
P._nigra_2              ACGTAATGAAATAAGATTGATTTATGGCTGGATTATCATTGGCTAACATA   [1180]
S._integra              ------------------------------------------------   [1007]
S._nigra                ------------------------------------------------   [516]
P._fremontii            ------------------------------------------------   [641]
P._angulata             ------------------------------------------------   [632]
P._laurifolia_1         ACGTAATGAAATAAGATTGATTTATGGCTGGATTATCATTGGCTAACATA   [1178]
P._laurifolia_2         ACGTAATGAAATAAGATTGATTTATGGCTGGATTATCATTGGCTAACATA   [1174]
P._simonii              ACGTAATGAAATAAGATTGATTTATGGCTGGAT-ATCAT-----------   [1163]
P._maximowiczii_1       ACGTAATGAAATAAGATTGATTTATGGCTGGAT-AT--------------   [1162]
P._maximowiczii_2       ACGTAATGAAATAAGATTGATTTATGGCTGGAT-A---------------   [1161]
P._deltoides_1          ACGTAATGAAATAAGATTGATTTATGGCTGG-------------------   [926]
P._sargentii            ACGTAATGAAATAAGATTGATTTATGGCTGGAT-ATCAT-----------   [1165]
P._deltoides_2          ACGTAATGAAATAAGATTGATTTATGGCTGGAT-A---------------   [1163]
P._deltoides_3          ACGTAATGAAATAAGATTGATTTATGGCTG--------------------   [1159]
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[                              1210       1220  ]
[                              .          .     ]

P._alba_1                  -------------------------   [564]
P._alba_3                  -------------------------   [1165]
P._alba_2                  -------------------------   [1165]
P._grandidentata_2         -------------------------   [1160]
P._grandidentata_3         -------------------------   [1158]
P._grandidentata_4         -------------------------   [1156]
P._davidiana_2             -------------------------   [1166]
P._davidiana_5             -------------------------   [1152]
P._davidiana_4             -------------------------   [744]
P._davidiana_3             -------------------------   [752]
P._tremula_1               -------------------------   [1156]
P._tremuloides_2           -------------------------   [1157]
P._davidiana_1             -------------------------   [1157]
P._tremuloides_1           -------------------------   [1144]
P._trichocarpa_1           -------------------------   [859]
P._balsamifera_1           -------------------------   [860]
P._balsamifera_2           -------------------------   [814]
P._cathayana               -------------------------   [706]
P._tristis                 -------------------------   [635]
P._angustifolia            -------------------------   [851]
P._trichocarpa_2           -------------------------   [873]
P._balsamifera_3           -------------------------   [808]
P._szechuanica             -------------------------   [1158]
P._nigra_3                 TGAAATAAGATTGATTTATGGCTGG   [1203]
P._nigra_5                 -------------------------   [818]
P._nigra_1                 -------------------------   [842]
P._nigra_4                 TGAAATAAGATTGATTTATGGCTGG   [1204]
P._nigra_2                 TGAAATAAGATTGATTTATGGCTG-   [1204]
S._integra                 -------------------------   [1007]
S._nigra                   -------------------------   [516]
P._fremontii               -------------------------   [641]
P._angulata                -------------------------   [632]
P._laurifolia_1            TGAAATAAGATTGATTTATGGCTGG   [1203]
P._laurifolia_2            TGAAATAAGATTGATTTATGGCTGG   [1199]
P._simonii                 -------------------------   [1163]
P._maximowiczii_1          -------------------------   [1162]
P._maximowiczii_2          -------------------------   [1161]
P._deltoides_1             -------------------------   [926]
P._sargentii               -------------------------   [1165]
P._deltoides_2             -------------------------   [1163]
P._deltoides_3             -------------------------   [1159]
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Appendix 2. 5 Nucleotide sequence data matrix of SCISSR fragment PIS8A2.

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[                          10         20         30         40         50]
[                          .          .          .          .          .]

P._cathayana               ---------------------------------------------AACC    [4]
P._simonii                 ----TGAAGACCTGTTAACGAATTTCTCCTCGAGCAAGAAACCAGTAACC    [46]
P._nigra_1                 ----TGAAGGCCTGTTAACGAATTTCTCCTCGAGCAAGAAACCAGTAACC    [46]
P._nigra_3                 ----TGAAGGCCTGTTAACGAATTTCTCCTCGAGCAAGAAACCAGTAACC    [46]
P._nigra_4                 -CAATGAAGGCCTGTTAACGAATTTCTCCTCGAGCAAGAAACCAGTAACC    [49]
P._nigra_5                 ACAATGAAGGCCTGTTAACGAATTTCTCCTCGAGCAAGAAACCAGTAACC    [50]
P._nigra_2                 ---ATGAAGGCCTGTTAACGAATTTCTCCTCGAGCAAGAAACCAGTAACC    [47]
P._angustifolia            ----------------------------------AAAAACCACTAACC    [14]
P._alba_1                  ---ATGAAGACCTGTTAACAAATTTCTCCTCGAGCGAGAAACCAGTAACC    [47]
P._alba_2                  ------------TGTTAACAAATTTCTCCTCGAGCGAGAAACCAGAAACC    [38]
P._alba_3                  ACA-TGAAGACCTGTTAACAAATTTCTCCTCGAGCGAGAAACCAGTAACC    [49]
P._tremuloides_1           -----GAAGACCTGTTAACAAATTTCTCCTCGAGCGAGAAACCAGTAACC    [45]
P._grandidentata_3         --AATGAAGACCTGTTAACAAATTTCTCCTCGAGCGAGAAACCAGTAACC    [48]
P._grandidentata_4         ---ATGAAGACCTGTTAACAAATTTCTCCTCGAGCGAGAAACCAGTAACC    [47]
P._davidiana_2             -------------GTTAACAAATTTCTCCTCGAGCGAGAAACCAGTAACC    [37]
P._davidiana_1             -CAATGAAGACCTGTTAACAAATTTCTCCTCGAGCGAGAAACCAGTAACC    [49]
P._davidiana_5             ----TGAAGACCTGTTAACAAATTTCTCCTCGAGCGAGAAACCAGTAACC    [46]
P._tremuloides_2           --A-TGAATACCTGTTAACTA-TTTCTCCTCGAGCGAGAAACCAGTAACC    [46]
P._tremula_1               -CAATGAAGACCTGTTAAAAA-TTTCTCCTCGAGCGAGAAACCAGTAACC    [48]
P._maximowiczii_1          -CAATGAAGACCTGTTAACGAATTTCTCCTCGAGCAAGAAACCAGTAACC    [49]
P._balsamifera_2           -CAATGAAGACCTGTTAACAAATTTCTCCTCGAGCAAGAAACCAGTAACC    [49]
P._laurifolia_2            ---ATGAAGACCTGTTAACGAATTTCTCCTCGAGCAAGAAACCAGTAACC    [47]
P._laurifolia_1            ---ATGAAGACCTGTTAACGAATTTCTCCTCGAGCAAGAAACCAGTAACC    [47]
P._trichocarpa_1           ----TGAAGACCTGTTAA-AAATTTCTCCTCGAGCAAGAAACCAGTAACC    [45]
P._trichocarpa_2           ----TGAAGACCTGTTAACAAATTTCTCCTCGAGCAAGAAACCAGTAACC    [46]
P._szechuanica             ---ATGAAGACCTGTTAACAAATTTCTCCTCGAGCAAGAAACCAGTAACC    [47]
P._deltoides_1             --------------------AATTTCTCCTCGAGCAAGAAACCAGTAACC    [30]
P._sargentii               ----TGAAGACCTGTTAA-AAATTTCTCCTCGAGCAAGAAACCAGTAACC    [45]
P._deltoides_2             ---ATGAAGACCTGTTAACAAATTTCTCCTCGAGCAAGAAACCAGTAACC    [47]
P._balsamifera_1           ----TGAAGACCTGTTAACAAATTTCTCCTCGAGCAAGAAACCACCAACC    [46]
S._integra                 ----------------------TTTCTCCTAAATCAAGAAGCCAGTAACC    [28]
S._nigra                   ---------------------------------------------------    [0]

[                          60         70         80         90        100]
[                          .          .          .          .          .]

P._cathayana               GCTGCTCTTTATTATCTAGGAAGAAAGAGAGGGGATATTCATATTCCT--    [52]
P._simonii                 GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [94]
P._nigra_1                 GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [94]
P._nigra_3                 GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [94]
P._nigra_4                 GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [97]
P._nigra_5                 GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [98]
P._nigra_2                 GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [95]
P._angustifolia            GCTGCTCTTTATTACACAGAAAAAAAGAGAGGGGATATTCTCATTCCC--    [62]
P._alba_1                  GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [95]
P._alba_2                  GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [86]
P._alba_3                  GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [97]
P._tremuloides_1           GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [93]
P._grandidentata_3         GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [96]
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P._grandidentata_4    GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [95]
P._davidiana_2        GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [85]
P._davidiana_1        GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [97]
P._davidiana_5        GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [94]
P._tremuloides_2      GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [94]
P._tremula_1          GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [96]
P._maximowiczii_1     GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [97]
P._balsamifera_2      GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [97]
P._laurifolia_2       GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [95]
P._laurifolia_1       GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [95]
P._trichocarpa_1      GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [93]
P._trichocarpa_2      GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [94]
P._szechuanica        GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [95]
P._deltoides_1        GCTGCTCTTTACTACAGGGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [78]
P._sargentii          GCTGCTCTTTACTACAGGGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [93]
P._deltoides_2        GCTGCTCTTTACTACAGGGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [95]
P._balsamifera_1      GCTGCTCTTTATTACAGAGGAAGAAAGAGAGGGGATATTCTTATTCCT--    [94]
S._integra            GCTGCTCTTTATTGCAGAGGAAGAAAGAGAGGGGATATTCATATTCCTAT    [78]
S._nigra              ------CTTTATGGCAAAGAAAAAAAAGAAGGGAATATCCTATTCCCTAT    [44]

[                              110       120       130       140       150]
[                              .         .         .         .         .]

P._cathayana          ------CATGGCTTGCTTTCGTGCGTGTGTGGGTTCATCCAAAGAACGCA    [96]
P._simonii            ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [138]
P._nigra_1            ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [138]
P._nigra_3            ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [138]
P._nigra_4            ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [141]
P._nigra_5            ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [142]
P._nigra_2            ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [139]
P._angustifolia       ------CACGGGGCGCTCTCTCGCGTGTGTTGGGTCACCCCAAAAACACA    [106]
P._alba_1             ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [139]
P._alba_2             ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [130]
P._alba_3             ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [141]
P._tremuloides_1      ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [137]
P._grandidentata_3    ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [140]
P._grandidentata_4    ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [139]
P._davidiana_2        ------CAGGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [129]
P._davidiana_1        ------CATGGCTTGCTTTCTTGCGTGTTTCGGTTCATCCAAAGAACGCA    [141]
P._davidiana_5        ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [138]
P._tremuloides_2      ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [138]
P._tremula_1          ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [140]
P._maximowiczii_1     ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [141]
P._balsamifera_2      ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [141]
P._laurifolia_2       ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [139]
P._laurifolia_1       ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [139]
P._trichocarpa_1      ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [137]
P._trichocarpa_2      ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [138]
P._szechuanica        ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [139]
P._deltoides_1        ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [122]
P._sargentii          ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [137]
P._deltoides_2        ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [139]
P._balsamifera_1      ------CATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [138]
S._integra            CTTGGATATGGCTTGCTTTCTTGCGTGTTTTGGTTCATCCAAAGAACGCA    [128]
S._nigra              CTGGAAAATGGTTGGCTTCCTGGCGGGTTTGGGTCCACCCAAGAAACCCA    [94]

[                              160       170       180       190       200]
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[                                .         .         .         .         .]

P._cathayana          AACGTCGTAGGCACAGGAAGGTTCAAGCACGAGCCCGTGTAAGAACCTTA   [146]
P._simonii            AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [188]
P._nigra_1            AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [188]
P._nigra_3            AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [188]
P._nigra_4            AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [191]
P._nigra_5            AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [192]
P._nigra_2            AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [189]
P._angustifolia       CACATCTTATGCACACTAAGGTGCACCCCCGAGTCCCTGTGAAAAACTCA   [156]
P._alba_1             AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [189]
P._alba_2             AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [180]
P._alba_3             AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [191]
P._tremuloides_1      AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [187]
P._grandidentata_3    AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [190]
P._grandidentata_4    AACGTCGTACGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [189]
P._davidiana_2        CACGTCGTACGCACAGTAGGGTTCAACCACGAGTCCGTGTAAAAACCTTA   [179]
P._davidiana_1        AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [191]
P._davidiana_5        AACGTCGTAGGCACAGTAAGGTGCAACCACGAGTCCATGTAAGAACCTTA   [188]
P._tremuloides_2      AACCTCGTACGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [188]
P._tremula_1          AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [190]
P._maximowiczii_1     NACGTCGTAGGCACAGTANGGTTCAACCACGAGTCCATGTAAGAACCTTA   [191]
P._balsamifera_2      NACGTCGTAGGCACAGTANGGTTCAACCACGAGTCCATGTAAGAACCTTA   [191]
P._laurifolia_2       AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [189]
P._laurifolia_1       AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [189]
P._trichocarpa_1      AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [187]
P._trichocarpa_2      AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [188]
P._szechuanica        AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [189]
P._deltoides_1        AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [172]
P._sargentii          AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [187]
P._deltoides_2        AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [189]
P._balsamifera_1      AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [188]
S._integra            AACGTCGTAGGCACAGTAAGGTTCAACCACGAGTCCATGTAAGAACCTTA   [178]
S._nigra              ACCTCCGAAGGACCATAAAGGTCCACCCCCAATCCCTGGTAAAACCCTTA   [144]

[                              210       220       230       240       250]
[                              .         .         .         .         .]

P._cathayana          AGGAGCCTGGTTTCCGAAAATTTCCCTTTTGGGTTTCGTTATTTTTT-CT   [195]
P._simonii            AGGAGCCTTGTTTCCTAAAATTTCCCTTTTGGGTTTCCTTATTTTTT-CT   [237]
P._nigra_1            AGGAGCCTTGTTTCCTAAAATTTCCCTTTTGGGTTTCGTTATTTTTTTCT   [238]
P._nigra_3            AGGAGCCTTGTTTCCTAAAATTTCCCTTTTGGGTTTCGTTATTTTTTTCT   [238]
P._nigra_4            AGGAGCCTTGTTTCCTAAAATTTCCCTTTTGGGTTTCGTTATTTTTTTCT   [241]
P._nigra_5            AGGAGCCTTGTTTCCTAAAATTTCCCTTTTGGGTTTCGTTATTTTTTTCT   [242]
P._nigra_2            AGGAGCCTTGTTTCCTAAAATTTCCCTTTTGGGTTTCGTTATTTTTTTCT   [239]
P._angustifolia       TAGAGCCTTGTGTCCCAAAATTTCCCTTTTGGGTTTCCTTATTTTTT-CT   [205]
P._alba_1             AGGAGCCTTGTTTCCTAAAATTTCCCTTTTGGGTTTCCTTATTTTTT-AT   [238]
P._alba_2             AGGAGCCTTGTTTCCTAAAATTTCCCTTTTGGGTTTCCTTATTTTTT-AT   [229]
P._alba_3             AGGAGCCTTGTTTCCTAAAATTTCCCTTTTGGGTTTCCTTATTTTTT-AT   [240]
P._tremuloides_1      GGGAGCCTTGTTTCCTAAAATTTCCCTTTTGGGTTTCCTTATTTTTT-AT   [236]
P._grandidentata_3    AGGAGCCTTGTTTCCTAAAATTTCCCTTTTGGGTTTCCTTATTTTTT-AT   [239]
P._grandidentata_4    AGGAGCCTTGTTTCCTAAAATTTCCCTTTTGGGTTTCCTTATTTTTT-AT   [238]
P._davidiana_2        GGGAGCCTTGTTTCCTAAAATTTCCCTTTTAGGTTTCCATATTTTTT-AT   [228]
P._davidiana_1        AGGAGCCTTGTTTCCTAAAATTTCCCTTTTAGGTTTCCTTATTTTTT-AT   [240]
P._davidiana_5        AGGAGCCTTGTTTCCTAAAATTTCCCTTTTAGGTTTCCTTATTTTTT-AT   [237]
P._tremuloides_2      GGGAGCCTTGTTTCCAAAAATTTCCCTTTTGGGTTTCCTTATTTTTT-AT   [237]
P._tremula_1          AGGAGCCTTGTTTCCTAAAATTTCCCTTTTAGGTTTCCTTATTTTTT-AT   [239]
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P._maximowiczii_1     AGGAGCCTTGTTTCCTAAAATTTCCCTTTTGGGTTTCCTTATTTTTT-CT   [240]
P._balsamifera_2      AGGAGCCTTGTTTCCTAAAATTTCCCTTTTGGGTTTCCTTATTTTTT-CT   [240]
P._laurifolia_2       AGGAGCCTTGTTTCCTAAAATTTCCCTTTTGGGTTTCGTTATTTTTT-CT   [238]
P._laurifolia_1       AGGAGCCTTGTTTCCTAAAATTTCCCTTTTGGGTTTCGTTATTTTTT-CT   [238]
P._trichocarpa_1      AGGAGCCTTGTTTCCTAAATTTTCCCTTTTGGGTTTCGTTATTTTTT-CT   [236]
P._trichocarpa_2      AGGAGCCTTGTTTCCTAAAATTTCCCTTTTGGGTTTCGTTATTTTTT-CT   [237]
P._szechuanica        AGGAGCCTTGTTTCCTAAATTTTCCCTTTTGGGTTTCGTTATTTTTT-CT   [238]
P._deltoides_1        AGGAGCCTTGTTTCCTAAAATTTCCCTTTTGGGTTTCGTTATTTTTT-CT   [221]
P._sargentii          AGGAGCCTTGTTTCCTAAAATTTCCCTTTTGGGTTTCGTTATTTTTT-CT   [236]
P._deltoides_2        AGGAGCCTTGTTTCCTAAAATTTCCCTCTTGGGTTTCGTTATTTTTT-CT   [238]
P._balsamifera_1      AGGAGCCTTGTTTCCTAAAATTTTTCTTTTGGGTTTCCTTATTTTTT-CT   [237]
S._integra            AGGAGCCTTGTTTCCTAAAATGTCCCTTTTGGGTTTTTTTATATGTTTCT   [228]
S._nigra              AGGA-CCCTGGTTCCCAAAAACTCCCCTTTGGGTTTTTTTATATGTTTCT   [193]

[                              260        270        280        290        300]
[                              .          .          .          .          .]

P._cathayana          GTCTCTCTGCTTGATGTTCATGAAAACAAAAAAAAAGAAAAAAATAAGAC   [245]
P._simonii            GTCTCTCTGCTTGATTTTCAAGAAAACAGAGAAGAAGAGAAGAGTAGGAC   [287]
P._nigra_1            GTCTCTCCGCTTGATTTTCAAGAAAACAGAGAAGAAGAGAAGAGTAGGAT   [288]
P._nigra_3            GTCTCTCCGCTTGATTTTCAAGAAAACAGAGAAGAAGAGAAGAGTAGGAT   [288]
P._nigra_4            GTCTCTCCGCTTGATTTTCAAGAAAACAGAGAAGAAGAGAAGAGTAGGAT   [291]
P._nigra_5            GTCTCTCCGCTTGATTTTCAAGAAAACAGAGAAGAAGAGAAGAGTAGGAT   [292]
P._nigra_2            GTCTCTCCGCTTGATTTTCAAGAAAACAGAGAAGAAGAGAAGAGTAGGAT   [289]
P._angustifolia       CTCTCTCTGCGTGTTTTTCACAAAAACACAGAAAAAAAGAAAAGAATGAC   [255]
P._alba_1             GTCTCTCTGCTTGATTTTCAAGAAAACAGAGAAAAAGAGAAGAGTAGGAC   [288]
P._alba_2             GTCTCTCTGCTTGATTTTCAAGAAAACAGAGAAAAAGAGAAGAGTAGGAC   [279]
P._alba_3             GTCTCTCTGCTTGATTTTCAAGAAAACAGAGAAAAAGAGAAGAGTAGGAC   [290]
P._tremuloides_1      GTCTCTCTGCTTGATTTTCAAGAAAACAGAGAAGAAGAGAAGAGTAGGAC   [286]
P._grandidentata_3    GTCTCTCTGCTTGATTTTCAAGAAAACAGAGAAGAAGAGAAGAGGAGGTC   [289]
P._grandidentata_4    GTCTCTCTGCTTGATTTTCAAGAAAACAGAGAAAAAGAGAAGAGGAGGTC   [288]
P._davidiana_2        GTCTCTCTGCTTGATTTTCAAGAAAACAGAAAAAAAGAGAAGAGGAGGTC   [278]
P._davidiana_1        GTCTCTCTGCTTGATTTTCAAGAAAACAGAGAAGAAGAGAAGAGTAGGAC   [290]
P._davidiana_5        GTCTCTCTGCTTGATTTTCAAGAAAACAGAGAAGAAGAGAAGAGGAGGTC   [287]
P._tremuloides_2      GTCTCTCTGCTTGATTTTCAAGAAAACAGAAAAGAAGAGAAGAGGAGGTT   [287]
P._tremula_1          GTCTCTCTGCTTGATTTTCAAGAAAACAGAGAAGAAGAGAAGAGGAGGTC   [289]
P._maximowiczii_1     NTCTCTCTGCTTGATTTTCAAGAAAACAGAGAAGAAGAGAAGAGTAGGAC   [290]
P._balsamifera_2      GTCTCTCTGCTTGATTTTCAAGAAAACAGAGAANAAGAGAAGAGTANGAC   [290]
P._laurifolia_2       GTCTCTCTGCTTGATGTTCAAGAAAACAGAGAAGAAGAGAAGAGTAGGAC   [288]
P._laurifolia_1       GTCTCTCTGCTTGATGTTCAAGAAAACAGAGAAGAAGAGAAGAGTAGGAC   [288]
P._trichocarpa_1      GTCTCTCTGCTTGATTTTCAAGAAAACAGAGAAGAAGAGAAGAGTAGGAC   [286]
P._trichocarpa_2      GTCTCTCTGCTTGATTTTCAAGAAAACAGAGAAGAAGAGAAGAGTAGGAC   [287]
P._szechuanica        GTCTCTCTGCTTGATTTTCAAGAAAACAGAGAAGAAGAGAAGAGTAGGAC   [288]
P._deltoides_1        GTCTCTCTGCTTGATTTTCAAGAAAACAGAGAAGAAGAGAAGAGTAGGAC   [271]
P._sargentii          GTCTCTCTGCTTGATTTTCAAGAAAACAGAGAAGAAGAGAAGAGTAGGAC   [286]
P._deltoides_2        GTCTCTCTGCTTGATTTTCAAGAAAACAGAGAAGAAGAGAAGAGTAGGAC   [288]
P._balsamifera_1      GTCTCTCTGCTTGATTTTCAAGAAAACAGAGAAGAAGAGAAGAGTATGAC   [287]
S._integra            ATCTCTCTGCTTGGTTT-CAAGAGAACAGAGAAGAGGA-----GTAGGAT   [272]
S._nigra              AACTCTCTGCTTGCTTTCCTGA-GAACCAAGAAAACGAA-----TAAGAT   [237]

[                              310        320        330        340        350]
[                              .          .          .          .          .]

P._cathayana          CAACATTTTTGGTTCTTTTTGTTTGAGAGGCATAGGATTTCTAGAATTT-   [294]
P._simonii            CAACATTTTTTGTTCTTTTTGTTTGAGAGGCTTTGTATTTCTTGAATTT-   [336]
P._nigra_1            CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGTATTTCTTGAATTT-   [337]
P._nigra_3            CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGTATTTCTTGAATTT-   [337]
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P._nigra_4              CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGTATTTCTTGAATTT-    [340]
P._nigra_5              CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGTATTTCTTGAATTT-    [341]
P._nigra_2              CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGTATTTCTTGAATTT-    [338]
P._angustifolia         CCCCATTTTTGGGTCTTTTTGTGTGAGAGACTTTGTGTATCTCGAATTT-    [304]
P._alba_1               CAACATTTTTGGTTCTTCTTGTTTGAGAGGCTTTGGATTTCTTGAATTT-    [337]
P._alba_2               CCACATTTTTGGTTCTTCTTGTTTGAGAGGCTTTGGATTTCTTGAATTT-    [328]
P._alba_3               CAACATTTTTGGTTCTTCTTGTTTGAGAGGCTTTGGATTTCTTGAATTT-    [339]
P._tremuloides_1        AAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGGATTTCTTGAAGTT-    [335]
P._grandidentata_3      CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGGATTTCTTGAATTT-    [338]
P._grandidentata_4      CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGGATTTCTTGAATTT-    [337]
P._davidiana_2          CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGGATTTCTTGAATTT-    [327]
P._davidiana_1          CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGGATTTCTTGAATTT-    [339]
P._davidiana_5          CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGGATTTCTTGAATTT-    [336]
P._tremuloides_2        CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGGATTTCTTGAATTT-    [336]
P._tremula_1            CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGGATTTCTTGAATTT-    [338]
P._maximowiczii_1       CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGNATTTCTTGAATTT-    [339]
P._balsamifera_2        CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGGATTTCTTGAATTT-    [339]
P._laurifolia_2         CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGGATTTCTTGAATTT-    [337]
P._laurifolia_1         CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGGATTTCTTGAATTT-    [337]
P._trichocarpa_1        CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGGATTTCTTGAATTT-    [335]
P._trichocarpa_2        CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGGATTTCTTGAATTT-    [336]
P._szechuanica          CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGGATTTCTTGAATTT-    [337]
P._deltoides_1          CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGGATTTCTTGAATTT-    [320]
P._sargentii            CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGGATTTCTTGAATTT-    [335]
P._deltoides_2          CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGGATTTCTTGAATTT-    [337]
P._balsamifera_1        CAACATTTTTGGTTCTTTTTGTTTGAGAGGCTTTGGATTTCTTGAATTT-    [336]
S._integra              CAACATTTTTTGTTCTTTTTGTTTGAGAGGCTTTGGATTTCTTGATTTTT    [322]
S._nigra                CAACATGTCCTGCTCTTTTTGTTCGAGAGGCTATAGATTTCTTGATTTTT    [287]

[                                360       370       380       390       400]
[                                .         .         .         .         .]

P._cathayana            ----------CCTTTGCTTGTTGTGGCAACAAATTGATA-GTGTTTAA    [331]
P._simonii              ----------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTAA    [373]
P._nigra_1              ----------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTAA    [374]
P._nigra_3              ----------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTAA    [374]
P._nigra_4              ----------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTAA    [377]
P._nigra_5              ----------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTAA    [378]
P._nigra_2              ----------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTAA    [375]
P._angustifolia         ----------CCCTTGCGTGTGGTGTCAACAAATTGGGA-GTGTGTAA    [341]
P._alba_1               ----------CCTTTGATTGTTGTGTCAACAAATTGGTAAGTGTTGAA    [375]
P._alba_2               ----------CCTTTGATTGTTGTGTCAACAAATTGGTAAGTGTTGAA    [366]
P._alba_3               ----------CCTTTGATTGTTGTGTCAACAAATTGGTAAGTGTTGAA    [377]
P._tremuloides_1        ----------CCTTTGATTGTTGTGTCAACAAATTGGTA-GTGTTTAA    [372]
P._grandidentata_3      ----------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTTA    [375]
P._grandidentata_4      ----------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTTA    [374]
P._davidiana_2          ----------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTAA    [364]
P._davidiana_1          ----------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTAA    [376]
P._davidiana_5          ----------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTCAA    [373]
P._tremuloides_2        ----------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GCGTTTAA    [373]
P._tremula_1            ----------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTAA    [375]
P._maximowiczii_1       ----------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTAA    [376]
P._balsamifera_2        ----------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTAA    [376]
P._laurifolia_2         ----------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTAA    [374]
P._laurifolia_1         ----------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTAA    [374]
P._trichocarpa_1        ----------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTAA    [372]
P._trichocarpa_2        ----------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTAA    [373]
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P._szechuanica          ------------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTAA   [374]
P._deltoides_1          ------------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTAA   [357]
P._sargentii            ------------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTAA   [372]
P._deltoides_2          ------------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTAA   [374]
P._balsamifera_1        ------------CCTTTGCTTGTTGTGTCAACAAATTGGTA-GTGTTTAA   [373]
S._integra              GTTCTTTGATGTCCTTTGCTTGCTGTGTCAACAAATTGGTA-GTGTTTAA   [371]
S._nigra                GTTATTTGATGTCCTTTGCTCGCTGTGTCAGCAAATTAGAAATGGTT-AA   [336]

[                                410       420       430       440       450]
[                                .         .         .         .         .]

P._cathayana            GTCTGATTTTGATTTGTGCTTTTTT-GTTGCAGAGAAAGAAAGGTTATGG   [380]
P._simonii              GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGTAGGTTATGG   [422]
P._nigra_1              GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGTAGGTTATGG   [423]
P._nigra_3              GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGTAGGTTATGG   [423]
P._nigra_4              GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGTAGGTTATGG   [426]
P._nigra_5              GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGTAGGTTATGG   [427]
P._nigra_2              GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGTAGGTTATGG   [424]
P._angustifolia         ATCTGATATTGAGATGTGATTTTTT-GTGGCACAGAAAGGTAAGTTATGG   [390]
P._alba_1               GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGAAGGTTATGG   [424]
P._alba_2               GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGAAGGTTATGG   [415]
P._alba_3               GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGAAGGTTATGG   [426]
P._tremuloides_1        GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGAAGGTTATGG   [421]
P._grandidentata_3      GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGAAGGTTATGG   [424]
P._grandidentata_4      GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGAAGGTTATGG   [423]
P._davidiana_2          GTCTGATTTTGAGTTGTGATTTTT--GTTGCAGAGAAAGGAAGGTTATGG   [412]
P._davidiana_1          GTCTGATTTTGAGTTGTGATTTTTGTGTCACAGAAAAGAGAGTGTTATGG   [426]
P._davidiana_5          GTCTGATTTTGAGTTGTGATTTTTG-GTGCAAAAAAAGGAAGGGTTATGG   [422]
P._tremuloides_2        CTCAGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGAATGTTATGG   [422]
P._tremula_1            GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGAAGGTTATGG   [424]
P._maximowiczii_1       GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGAAGGTTGTGG   [425]
P._balsamifera_2        GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGANGGTTATGG   [425]
P._laurifolia_2         GTCTGATTTTGAGTTGTGCTTTTTT-GTTGCAGAGAAAGGAAGGTTATGG   [423]
P._laurifolia_1         GTCTGATTTTGAGTTGTGCTTTTTT-GTTGCAGAGAAAGGAAGGTTATGG   [423]
P._trichocarpa_1        GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGAAGGTTATGG   [421]
P._trichocarpa_2        GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGAAGGTTATGG   [422]
P._szechuanica          GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGAAGGTTATGG   [423]
P._deltoides_1          GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGAAGGTTATGG   [406]
P._sargentii            GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGAAGGTTATGG   [421]
P._deltoides_2          GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGAAGGTTATGG   [423]
P._balsamifera_1        GTCTGATTTTGAGTTGTGATTTTTT-GTTGCAGAGAAAGGTAGGTTATGG   [422]
S._integra              GTTTGATTTTGAGTTGTGATTTTT--GTTGCAGAGAAAGGAAGGTTATGG   [419]
S._nigra                GGTTGAATTTGAATTGTGATTTTTT-GTTGCTTAGAAAGGAAAGATATGG   [385]

[                                460       470       480       490       500]
[                                .         .         .         .         .]

P._cathayana            GAGTCCAATGCAAGCTACCGTTTCAGTGGTTAAGGACTGCTGCCCATACA   [430]
P._simonii              GAGTCCAGTGGAAGCTACAGTTTCAGTGGTTCAGGACTGCTGCCCAGAGA   [472]
P._nigra_1              GAGTCCAGTGGAAGCTACAGTTTCAGTGGTTCAGGACTGCAGCCCAGAGA   [473]
P._nigra_3              GAGTCCAGTGGAAGCTACAGTTTCAGTGGTTCAGGACTGCAGCCCAGAGA   [473]
P._nigra_4              GAGTCCAGTGGAAGCTACAGTTTCAGTGGTTCAGGACTGCAGCCCAGAGA   [476]
P._nigra_5              GAGTCCAGTGGAAGCTACAGTTTCAGTGGTTCAGGACTGCAGCCCAGAGA   [477]
P._nigra_2              GAGTCCAGTGGAAGCTACAGTTTCAGTGGTTCAGGACTGCAGCCCAGAGA   [474]
P._angustifolia         GAGACCCCTGGAAACTACACTTTCTCTGGGTCAAGAGTGCTCCCCACAGA   [440]
P._alba_1               TAATCCAGTGGAAGCTACCGTTTCAGTGGTTAAGGACTGCTGCCCAGAGA   [474]
P._alba_2               TAGTCCAGTGGAAGCTACCGTTTCAGTGGTTAAGGACTGCTGCCCTGAGA   [465]
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P._alba_3               TAATCCAGTGGAAGCTACCGTTTCAGTGGTTAAGGACTGCTGCCCTGAGA   [476]
P._tremuloides_1        TAGTCCAGTGGAAGCTACCGTTTCAGTGGTTAAGGACTGCTGCCCAGAGA   [471]
P._grandidentata_3      TAGTCCAGTGGAAGCTACCGTTTCAGTGGTTAATGACTGCTGCCCAGAGA   [474]
P._grandidentata_4      TAGTCCAGTGGAAGCTACCGTTTCAGTGGTTAATGACTGCTGCCCAGAGA   [473]
P._davidiana_2          TACTCCAGTGGAAGCTACCGTTTCAGTGGTTAAAGACTGCTGCCCAGAGA   [462]
P._davidiana_1          TACTCCAGTGAAATCCACCTTTTCTGTGGTAAGAGACCGCCGCACAGAAA   [476]
P._davidiana_5          AATCCCA-TGGAAACCTCCCTTTTCATGGTTAAAGAACGGCTCCCAAAAA   [471]
P._tremuloides_2        TACTCCAGTGGAATCTACCGTTTCAGTGGTTAAGGACTGCTGCCCAGAAA   [472]
P._tremula_1            TAGTCCAGTGGAAGCTACCGTTTCAGTGGTTAAGGACTGCTGCCCAGAAA   [474]
P._maximowiczii_1       GAGTCCAGTGGAAGCTACCGTTTCAGTGGTTNAGGACTGCTGCCCAGAGA   [475]
P._balsamifera_2        GAGTCCAGTGGAAGCTACCGTTTCAGTGGTTAAGGACTGCTGCCCNGAGA   [475]
P._laurifolia_2         GAGTCCAGTGGAAGCTACCGTTTCAGTGGTTAAGGACTGCTGCCCAGAGA   [473]
P._laurifolia_1         GAGTCCAGTGGAAGCTACCGTTTCAGTGGTTAAGGACTGCTGCCCAGAGA   [473]
P._trichocarpa_1        GAGTCCAGTGGAAGCTACCGTTTCAGTGGTTAAGGACTGCTGCCCAGAGA   [471]
P._trichocarpa_2        GAGTCCAGTGGAAGCTACCGTTTCAGTGGTTAAGGACTGCTGCCCTGAAA   [472]
P._szechuanica          GAGTCCAGTGGAAGCTACCGTTTCAGTGGTTAAGGACTGCTGCCCAGAGA   [473]
P._deltoides_1          TAGTCCAGTAGAAGCTACCGTTTCAGTGGTTAAGGACTGCTGCCCAGAGA   [456]
P._sargentii            TAGTCCAGTAGAAGCTTCCGTTTCAGTGGTTAAGGACTGCTGCCCAGAGA   [471]
P._deltoides_2          TAGTCCAGTAGAAGCTTCCGTTTCAGTGGTTAAGGACTGCTGCCCAGAGA   [473]
P._balsamifera_1        GAGTCCAGTGGAAGCTACCGTTTCAGTGGTTAAGGACTGCTGCCCAGAGA   [472]
S._integra              TAGTCCAGTGGAATCTACCGTTTCAGTGGTTAAGGACTGCTGCCCAGAGA   [469]
S._nigra                GAGTCCAGTGGAATCTACAGTTTCCCTGGTTAAGGACTGCTGCCCACAGA   [435]

[                                510       520       530       540       550]
[                                .         .         .         .         .]

P._cathayana            AACATATTGTGAGCCCAACTTCAGATATCAAGTTAGTAAGTAAAGGACTG   [480]
P._simonii              AGCCTATTGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAGGACTG   [522]
P._nigra_1              AGCCTATTGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAGGACTG   [523]
P._nigra_3              AGCCTATTGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAGGACTG   [523]
P._nigra_4              AGCCTATTGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAGGACTG   [526]
P._nigra_5              AGCCTATTGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAGGACTG   [527]
P._nigra_2              AGCCTATTGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAGGACTG   [524]
P._angustifolia         AACCTATTGTGAGCCCCCCTTCACAAATCTCGTGATAAAAAAAAAAGAGTG   [490]
P._alba_1               AGCCTATAGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAGGACTG   [524]
P._alba_2               AGCCTATAGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAGGACTG   [515]
P._alba_3               AGCCTATAGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAGGACTG   [526]
P._tremuloides_1        AGCCTATAGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAGGACTG   [521]
P._grandidentata_3      AGCCTATAGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAGGACTG   [524]
P._grandidentata_4      AGCCTATAGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAGGACTG   [523]
P._davidiana_2          AGCCTATAGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAGGACTG   [512]
P._davidiana_1          ACCCTATAGTGCCCCCATCTTCAAAAATCGTGTTAATAAATAAGAGACTG   [526]
P._davidiana_5          AACCTAATGTGGACCCCACTTCCAAAATTCAGGTAATAAATTAAGGAATG   [521]
P._tremuloides_2        AGCCTATAGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAGGACTG   [522]
P._tremula_1            AGCCTATAGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAGGACTG   [524]
P._maximowiczii_1       AGNCTATTGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAGGACTG   [525]
P._balsamifera_2        AGCCTATTGTGAGCCCAGCTTCAGAAATCAGGTTAGTA----------     [513]
P._laurifolia_2         AGCCTATTGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAGGACTG   [523]
P._laurifolia_1         AGCCTATTGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAGGACTG   [523]
P._trichocarpa_1        AGCCTATTGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAAGACTG   [521]
P._trichocarpa_2        AGCCTATTGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAAGACTG   [522]
P._szechuanica          AGCCTATTGTGAGCCCAGCTTCAGAAATCAGGTTAGTAAGTAAAAGACTG   [523]
P._deltoides_1          AGCCTATTGTGAGCCCAACTTCAGAAATCAGGTTAGTAAGTAAAGGATTG   [506]
P._sargentii            AGCCTATTGTGAGCCCAACTTCAGAAATCAGGTTAGTAAGTAAAGGATTG   [521]
P._deltoides_2          AGCCTATTGTGAGCCCAACTTCAGAAATCAGGTTAGTAAGTAAAGGATTG   [523]
P._balsamifera_1        AGCCTATTGTGAGCCCAGCTTCAGAAATCAGGTTAGTA----------     [510]
S._integra              AGCCTATTGTGAGCCCAGTTTCAGATATCAGGTTAGTAAGTAAAGGACTG   [519]
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S._nigra              ACCCTAAAGT---------CTCTGATGTCCCCCATTTCAGAAAAGGACTG   [476]

[                              560       570       580       590       600]
[                              .         .         .         .         .]

P._cathayana          TACGGGTTCGGAGG--TTTGCTGCTTTTCTTTTCTTTCCGTTTGAGTATC   [528]
P._simonii            TACGGTTTCTGAGGTTTTTGCTGCTTTTCTTTTCTGTTCGTTTTAGTATC   [572]
P._nigra_1            TACGGTTTCTGAGGTTTTTGCTGCTTTTCTTTTCTGTTCGTTTTAGTATC   [573]
P._nigra_3            TACGGTTTCTGAGGTTTTTGCTGCTTTTCTTTTCTGTTCGTTTTAGTATC   [573]
P._nigra_4            TACGGTTTCTGAGGTTTTTGCTGCTTTTCTTTTCTGTTCGTTTTAGTATC   [576]
P._nigra_5            TACGGTTTCTGAGGTTTTTGCTGCTTTTCTTTTCTGTTCGTTTTAGTATC   [577]
P._nigra_2            TACGGTTTCTGAGGTTTTTGCTGCTTTTCTTTTCTGTTCGTTTTAGTATC   [574]
P._angustifolia       TGCAGTTTCTGAGATTTTTGCTGCTTTTCTTTTCTTTCCGTTTTAGTATC   [540]
P._alba_1             TACGGTTTCTGAGG--TTTGCTGCTTTTCTTTTCTTTCCCTTTTAGTATC   [572]
P._alba_2             TACGGTTTATGAGG--TTTGCTGCTTTTCTTTTCTTTCCCTTTTAGTATC   [563]
P._alba_3             TACGGTTTATGAGG--TTTGCTGCTTTTCTTTTCTTTCCCTTTTAGTATC   [574]
P._tremuloides_1      TACGGTTTCTGAGG--TTTGCTGCTTTTCTTTTCTTTCCCTTTTAGTATC   [569]
P._grandidentata_3    TACGGTTTCTGAGG--TTTGCTGCTTTTTTTTTCTTTCCCTTTTAGTATC   [572]
P._grandidentata_4    TACGGTTTCTGAGG--TTTGCTGCTTTTTTTTTCTTTCCCTTTTAGTATC   [571]
P._davidiana_2        TACGGTTTCTGAGG--TTTGCTGCTTTTCTTTTCTTTCCCTTTTAGTATC   [560]
P._davidiana_1        CACTGTCTCAGAGG--TGCGCCGCTTCTCTTCTCTCCCCCTTAGAATATC   [574]
P._davidiana_5        GTCCGGTTCCGAAG--GTTGGCGGTTTTTCTTTCCTTCCCTTTTAATACC   [569]
P._tremuloides_2      TACGGTTTCAGAGG--TTTGCTGCTTTTCTTTTCTTTCCCTTTTAATATC   [570]
P._tremula_1          TACGGTTTCTGAGG--TTTGCTGCTTTTCTTTTCTTTCCCTTTTAGTATC   [572]
P._maximowiczii_1     TACGGTTTCTGAGG--TTT------------------------------   [542]
P._balsamifera_2      --------------------------------------------------   [513]
P._laurifolia_2       TACGGTTTCTGAGG--TTTGCTGCTTTTCTTTTCTTTCCGTTTTAGTATC   [571]
P._laurifolia_1       TACGGTTTCTGAGG--TTTGCTGCTTTTCTTTTCTTTCCGTTTTAGTATC   [571]
P._trichocarpa_1      TACGGTTTCTGAGG--TTTGCTGCTTTTCTTTTCTTTCCGTTTTAGTATC   [569]
P._trichocarpa_2      TACGGTTTCTGAGG--TTTGCTGCTTTTCTTTTCTTTCCGTTTTAGTATC   [570]
P._szechuanica        TACGGTTTCTGAGG--TTTGCTGCTTTTCTTTTCTTTCCGTTTTAGTATC   [571]
P._deltoides_1        TACTGTTTCTGAGG--TTTGCTGCTTTTCTTTTCTTTCCGTTTTAGTATC   [554]
P._sargentii          TACTGTTTCTGAGG--TTTGCTGCTTTTCTTTTCTTTCCGTTTTAGTATC   [569]
P._deltoides_2        TACTGTTTCTGAGG--TTTGCTGCTTTTCTTTTCTTTCCGTTTTAGTATC   [571]
P._balsamifera_1      --CGGTTTCTGAGG--TTTGCTGCTTTTCTTTTCTTTTCGTTTTAGTATC   [556]
S._integra            AGAGGTTTAC------TGCTTTTCTTTTCTTTTCTTTCCTTGTTAGTATC   [563]
S._nigra              AGAGGATTAAGGGG--CTTAAAGCTTTCCCGGTTTT--------------   [510]

[                              610       620       630       640       650]
[                              .         .         .         .         .]

P._cathayana          TGTTTTATGCAACTGTCAGAAGGGGAAAAAAGGGGTTGTGTGAATGGTTT   [578]
P._simonii            TGTTTTATGCAGCTTTCTGAAGGGGGAAAAGAGGGTTTTGTGCATGGTTT   [622]
P._nigra_1            TGTTTTATGCAGCTTTCTGAAGGGGGAAAAGAGGGTTTTGTGCATGGTTT   [623]
P._nigra_3            TGTTTTATGCAGCTTTCTGAAGGGGGAAAAGAGGGTTTTGTGCATGGTTT   [623]
P._nigra_4            TGTTTTATGCAGCTTTCTGAAGGGGGAAAAGAGGGTTTTGTGCATGGTTT   [626]
P._nigra_5            TGTTTTATGCAGCTTTCTGAAGGGGGAAAAGAGGGTTTTGTGCATGGTTT   [627]
P._nigra_2            TGTTTTATGCAGCTTTCTGAAGGGGGAAAAGAGGGTTTTGTGCATGGTTT   [624]
P._angustifolia       TGTTTTATGCAGCTCTCTGAAGGGGAAAAAAAGGGTTTTGTGAATGGTTT   [590]
P._alba_1             TGTTTTATGCAACTTTCTGAAGGGGAAAAAC-GGGTTTTGTGAATGGTTT   [621]
P._alba_2             TGTCTTATGCAACTTTCTGAAGGGGAAAAAC-GGGTTTTGTGAATGGTTT   [612]
P._alba_3             TGTTTTATGCAACTTTCTGAAGGGGAAAAAC-GGGTTTTGTGAATGGTTT   [623]
P._tremuloides_1      TGTTTTATGCAACTTTCTGAAGGGGAAAAAA-GGGTTTTGTGAATGGTTT   [618]
P._grandidentata_3    TGTTTTATGCAACTTTCTGAAGGGTAAAAAC-GGGTTTTGTGAATGGTTT   [621]
P._grandidentata_4    TGTTTTATGCAACTTTCTGAAGGGTAAAAAC-GGGTTTTGTGAATGGTTT   [620]
P._davidiana_2        TGTTTTATGCAACTTTCTGAAGGGGAAAAAC-GGGTTTTGTGAATGGTTT   [609]
P._davidiana_1        TGTTATACGCATCTCTCAGAGGGAAAAAAAC-GGGTTGTGAGAATGGTTT   [623]
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```
P._davidiana_5        CGGTTTAAGCCACTTTTCGAAAGGGAAAAAG-GGGTTT------------   [606]
P._tremuloides_2      TGTTTTATGCAACTTTCTGAAGGGGAAAAAC-GGGTTTTGTGAATGGTTT   [619]
P._tremula_1          TGTTTTATGCAACTTTCTGAAGGGGAAAAAC-GGGTTTTGTGAATGGTTT   [621]
P._maximowiczii_1     ------------------------------------------------   [542]
P._balsamifera_2      ------------------------------------------------   [513]
P._laurifolia_2       TGTTTTATGCAGCTTTCTGAAGGGGAAAAAAAGGGTTTTGTGAATGGTTT   [621]
P._laurifolia_1       TGTTTTATGCAGCTTTCTGAAGGGGAAAAAAAGGGTTTTGTGAATGGTTT   [621]
P._trichocarpa_1      TGTTTTATGCAGCTTTCTGAAGGGGAAAAAAAGGGTTTTGTGAATGGTTT   [619]
P._trichocarpa_2      TGTTTTATGCAGCTTTCTGAAGGGGAAAAAAAGGGTTTTGTGAATGGTTT   [620]
P._szechuanica        TGTTTTATGCAGCTTTCTGAAGGGGAAAAAAAGGGTTTTGTGAATGGTTT   [621]
P._deltoides_1        TGTTTTATGCAGCTTTCTGAAGGGGAAAAAAAGGGTTTTGTGAATGGTTT   [604]
P._sargentii          TGTTTTATGCAGCTTTCTGAAGGGGAAAAAAAGGGTTTTGTGAATGGTTT   [619]
P._deltoides_2        TGTTTTATGCAGCTTTCTGAAGGGGAAAAAAAGGGTTTTGTGAATGGTTT   [621]
P._balsamifera_1      TGTTTTATGCAGCTTTCTGAAGGGGAAAAAAAGGGTTTTGTGAATGGTTT   [606]
S._integra            TGTCTTATGCAGCTTTCTGAAGGGGAGAAAA-GGGTTTTGTTAACGGTTT   [612]
S._nigra              ------------------------------------------------   [510]

[                              660        670        680        690        700]
[                              .          .          .          .          .]

P._cathayana          TTGAATTGCTGTATTTTGGGTTTTTCATGGGTGTATAGGGAT--------   [620]
P._simonii            TTGAATTGCTGTATTTTGGGTTTTTCATGGTTGTATAGGGATGATGGGTC   [672]
P._nigra_1            TTGAATTGCTGTATTTTGGGTTTTTCATGGTTGTATAGGGATGATGGGTC   [673]
P._nigra_3            TTGAATTGCTGTATTTTGGGTTTTTCATGGTTGTATAGGGATGATGGGTC   [673]
P._nigra_4            TTGAATTGCTGTATTTTGGGTTTTTCATGGTTGTATAGGGATGATGGGTC   [676]
P._nigra_5            TTGAATTGCTGTATTTTGGGTTTTTCATGGTTGTATAGGGATGATGGGTC   [677]
P._nigra_2            TTGAATTGCTGTATTTTGGGTTTTTCATGGTTGTATAGGGATGATGGGTC   [674]
P._angustifolia       TTGAATTGCTGTATTTTGGGTTTTTCATGGTTGTATAGGGATGATGGGTC   [640]
P._alba_1             TTGAATTGCTGTATTTTGGGTTTCTCATGGTTGTATAGGGATGATGGGTC   [671]
P._alba_2             TTGAATTGCTGTATTTTGGGTTTCTCATGGTTGTATAGGGATGATGGGTC   [662]
P._alba_3             TTGAATTGCTGTATTTTGGGTTTCTCATGGTTGTATAGGGATGATGGGTC   [673]
P._tremuloides_1      TTGAATTGCTGTATTTTGGGTTTCTCATGGTTGTATAGGGATGATGGGTC   [668]
P._grandidentata_3    TTGAATTGCTGTATTTTGGGTTTCTCATGGTTGTATAGGGATGATGGGTC   [671]
P._grandidentata_4    TTGAATTGCTGTATTTTGGGTTTCTCATGGTTGTATAGGGATGATGGGTC   [670]
P._davidiana_2        TTGAATTGCTGTATTTTGGGTTTCTCATGGTTGTATAGGGATGATGGGTC   [659]
P._davidiana_1        TAAAATCGCTGTATT-----------------------------------   [638]
P._davidiana_5        ------------------------------------------------   [606]
P._tremuloides_2      TTGAATTGCTGTATTTT---------------------------------   [636]
P._tremula_1          TTGAATTGCTGTATTTTGGGTTTCTCATGGTTGTATAGGGATGATGGGTC   [671]
P._maximowiczii_1     ------------------------------------------------   [542]
P._balsamifera_2      ------------------------------------------------   [513]
P._laurifolia_2       TTGAATTGCTGTATTTTGGGTTTTTCATGGTTGTATAGGGATGATGGGTC   [671]
P._laurifolia_1       TTGAATTGCTGTATTTTGGGTTTTTCATGGTTGTATAGGGATGATGGGTC   [671]
P._trichocarpa_1      TTGAATTGCTGTATTTTGGGTTTTTCATGGTTGTATAGGGATGATGGGTC   [669]
P._trichocarpa_2      TTGAATTGCTGTATTTTGGGTTTTTCATGGTTGTATAGGGATGATGGGTC   [670]
P._szechuanica        TTGAATTGCTGTATTTTGGGTTTTTCATGGTTGTATAGGGATGATGGGTC   [671]
P._deltoides_1        TTGAATTGCTGTATTTTGGGTTTTTCATGGTTGTATAGGGATGATGGGTC   [654]
P._sargentii          TTGAATTGCTGTATTTTGGGTTTTTCATGGTTGTATAGGGATGATGGGTC   [669]
P._deltoides_2        TTGAATTGCTGTATTTTGGGTTTTTCATGGTTGTATAGGGATGATGGGTC   [671]
P._balsamifera_1      TTGAATTGCTGTATTTTGGGTTTTTCATGGTTGTATAGGGATGATGGGTC   [656]
S._integra            TTGAATTGTTGCATTTTGGGTTTTCCATGGTCGTATAGGGATGATGGGTC   [662]
S._nigra              ------------------------------------------------   [510]

[                              710        720        730        740        750]
[                              .          .          .          .          .]

P._cathayana          ------------------------------------------------   [620]
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P._simonii              TGAGGAGAAATTAAGCTTGAGTACGAGAAAGAAAGTGACATTCAATTCGA   [722]
P._nigra_1              TGAGGAGAAATTAAGCTTGAGTACGAGAAAGAAAGTGACATTCAATTCGA   [723]
P._nigra_3              TGAGGAGAAATTAAGCTTGAGTACGAGAAAGAAAGTGACATTCAATTCGA   [723]
P._nigra_4              TGAGGAGAAATTAAGCTTGAGTACGAGAAAGAAAGTGACATTCAATTCGA   [726]
P._nigra_5              TGAGGAGAAATTAAGCTTGAGTACGAGAAAGAAAGTGACATTCAATTCGA   [727]
P._nigra_2              TGAGGAGAAATTAAGCTTGAGTACGAGAAAGAAAGTGACATTCAATTCGA   [724]
P._angustifolia         --------------------------------------------------   [640]
P._alba_1               TGAGGAGAAATTAAGCTTGAGTACGAGAAAGAAAGTGACATTCAATTCGA   [721]
P._alba_2               TGAGGAGAAATTAAGCTTGAGTACGAGAAAGAAAGTGACATTCAATTCGA   [712]
P._alba_3               TGAGGAGAAATTAAGCTTGAGTACGAGAAAGAAAGTGACATTCAATTCGA   [723]
P._tremuloides_1        TGAGGAGAAATTAAGCTTGAGTACGAGAAAGAAAGTGACATTCAATTCGA   [718]
P._grandidentata_3      TGAGGAAAAATTAAGCTTGAGTACGAGAAAGAAAGTGACATTCAATTCGA   [721]
P._grandidentata_4      --------------------------------------------------   [670]
P._davidiana_2          TG----------------------------------------------   [661]
P._davidiana_1          --------------------------------------------------   [638]
P._davidiana_5          --------------------------------------------------   [606]
P._tremuloides_2        --------------------------------------------------   [636]
P._tremula_1            TGAGGAGAAATTAAGCTTGAGTACGAGAAAGAAAGTGACATTCAATTCGA   [721]
P._maximowiczii_1       --------------------------------------------------   [542]
P._balsamifera_2        --------------------------------------------------   [513]
P._laurifolia_2         TGAGGAGAAATTAAGCTTGAGTACGAGAAAGAAAGTGACATTCAATTCGA   [721]
P._laurifolia_1         TGAGGAGAAATTAAGCTTGAGTACGAGAAAGAAAGTGACATTCAATTCGA   [721]
P._trichocarpa_1        TGAGGAGAAATTAATCTTGAGTACGAGAAAGAAAGTGACATTCAATTCGA   [719]
P._trichocarpa_2        TGAGGAGAAATTAATCTTGAGTACGAAAAAGAAAGTGACTTTCAATTCGA   [720]
P._szechuanica          TGAGGAGAAATTAAGCTTGAGTACGAGAAAGAAAGTGACATTCAATTCGA   [721]
P._deltoides_1          TGAGGAGAAATTAAGCTTGAGTACGAGAAAGAAAGTGACATTCAATTCGA   [704]
P._sargentii            TGAGGAGAAATTAAGCTTGAGTACGAGAAAGAAAGTGACATTCAATTCGA   [719]
P._deltoides_2          TGAGGAGAAATTAAGCTTGAGTACGAGAAAGAAAGTGACATTCAATTCGA   [721]
P._balsamifera_1        TGAGGAGAAATTAAGCTTGAGTACGAGAAAGAAAGTGACATTCAATTCGA   [706]
S._integra              TGAGGAGAAATTGAGCTTGAGTACGAGAAAGAAAGTGACATTTAATTCGA   [712]
S._nigra                --------------------------------------------------   [510]

[                              760       770       780       790       800]
[                              .         .         .         .         .]

P._cathayana            --------------------------------------------------   [620]
P._simonii              ATGTGACAACCTACGATCATGTTTCAGTTGAAGAATCCACAGATTTTACT   [772]
P._nigra_1              ATGTGACAACCTACGATCATGTTTCAGTTGAAGAATCCACAGATTTTACT   [773]
P._nigra_3              ATGTGACAACCTACGATCATGTTTCAGTTGAAGAATCCACAGATTTTACT   [773]
P._nigra_4              ATGTGACAACCTACGATCATGTTTCAGTTGA------------------   [757]
P._nigra_5              ATGTGACAACCTACGATCATGTTTCAGTTGAAGAATCCACAGATTTTACT   [777]
P._nigra_2              ATGTGACAACCTACGATCATGTTTCAGTTGAAGAATCCACAGATTTTACT   [774]
P._angustifolia         --------------------------------------------------   [640]
P._alba_1               ATGTGACAACCTACGACCATGTTTCAGTTGAAGAATCCACAGATTTTACG   [771]
P._alba_2               ATGTGACAACCTACGACCATGTTTCAGTTGAAGAATCCACAGATTTTACG   [762]
P._alba_3               ATGTGACAA---------------------------------------   [732]
P._tremuloides_1        ATGTGACAACCTACGA--------------------------------   [734]
P._grandidentata_3      ATGTGACAACCTACGACCATGTTTC-----------------------   [746]
P._grandidentata_4      --------------------------------------------------   [670]
P._davidiana_2          --------------------------------------------------   [661]
P._davidiana_1          --------------------------------------------------   [638]
P._davidiana_5          --------------------------------------------------   [606]
P._tremuloides_2        --------------------------------------------------   [636]
P._tremula_1            ATGTGACAACCTAC----------------------------------   [735]
P._maximowiczii_1       --------------------------------------------------   [542]
P._balsamifera_2        --------------------------------------------------   [513]
P._laurifolia_2         ATGTGACAACCTACGATCATGTTTCA----------------------   [747]
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P._laurifolia_1      ATGTGACAACCTACGATCATGTTTCAGTTGAAGAATCCACATATTTTACT   [771]
P._trichocarpa_1     ATGTGACAACCTACGATCATGTTTCAGTTGAAGAATCCTCAGATTTTACT   [769]
P._trichocarpa_2     ATGTGACAACCTACGATCATGTTTC------------------------   [745]
P._szechuanica       ATGTGACAACCTACGATCATGTTTC------------------------   [746]
P._deltoides_1       ATGTGACAACCTACGATCATGTTTCAGTTGAAGAATCCACAGATTTTACT   [754]
P._sargentii         ATGTGACAACCTACGATCATGTTTCAGTTGAAGAATCCACAGATTTTACT   [769]
P._deltoides_2       ATGTGACAACCTACGATCATGTTTC------------------------   [746]
P._balsamifera_1     ATGTGACAACCTACGATCATGTTTCAGTTGAAGAATCCTCAGATTTTACT   [756]
S._integra           ATGTGACAACCTACGACCATGTTTCAGTTGAAGAATCCACAGATTTTACG   [762]
S._nigra             --------------------------------------------------   [510]

[                             810       820       830       840       850]
[                             .         .         .         .         .]

P._cathayana         --------------------------------------------------   [620]
P._simonii           TTGGGGAAAGAAGATTGTGGCGATAAAAGGGAGGGAAAGGAGGAGAATAT   [822]
P._nigra_1           TTGGGGAAAGAAGATTGTGGCGATAAAAGGGAGGGAAAGGA---------   [814]
P._nigra_3           TTGGGG--------------------------------------------   [779]
P._nigra_4           --------------------------------------------------   [757]
P._nigra_5           TTGG----------------------------------------------   [781]
P._nigra_2           TTGGG---------------------------------------------   [779]
P._angustifolia      --------------------------------------------------   [640]
P._alba_1            TTGGGGAAAGAAGATTGTGGCGATAAAAGGGAGGGAAATGAGGAGAATAT   [821]
P._alba_2            TTGGGGAAAGAAGATTGTGGCGATAAAAGGGAGGGAAATGAGGAGAATAT   [812]
P._alba_3            --------------------------------------------------   [732]
P._tremuloides_1     --------------------------------------------------   [734]
P._grandidentata_3   --------------------------------------------------   [746]
P._grandidentata_4   --------------------------------------------------   [670]
P._davidiana_2       --------------------------------------------------   [661]
P._davidiana_1       --------------------------------------------------   [638]
P._davidiana_5       --------------------------------------------------   [606]
P._tremuloides_2     --------------------------------------------------   [636]
P._tremula_1         --------------------------------------------------   [735]
P._maximowiczii_1    --------------------------------------------------   [542]
P._balsamifera_2     --------------------------------------------------   [513]
P._laurifolia_2      --------------------------------------------------   [747]
P._laurifolia_1      TTGGGG--------------------------------------------   [777]
P._trichocarpa_1     TTGGGGAAAGAAGATTGTGGCGATAAAAGGGAGGGAAAGGAGGAGAATAT   [819]
P._trichocarpa_2     --------------------------------------------------   [745]
P._szechuanica       --------------------------------------------------   [746]
P._deltoides_1       TTGGGGAAAGAAGATTGTGGCGATAAAAGGGAGGGAAAGGAGGAGAATAT   [804]
P._sargentii         TTGGGGAAAGAAGATTGTGGCGATAAAAGGGAGGGAAAGGAGGAGAATAT   [819]
P._deltoides_2       --------------------------------------------------   [746]
P._balsamifera_1     TTGGGGAAAGAAGATTGTGGCGATAAAAGGGAGGGAAAGGAGGAGAACAT   [806]
S._integra           TTGGGTAAAGAAGATGGTGGCGATAAAAGGGAGGGAAAGGAGGAGAATAG   [812]
S._nigra             --------------------------------------------------   [510]

[                             860       870       880       890       900]
[                             .         .         .         .         .]

P._cathayana         --------------------------------------------------   [620]
P._simonii           TGCAAAACCAAGCCAATCTCAGTCTTCTT---------------------   [851]
P._nigra_1           --------------------------------------------------   [814]
P._nigra_3           --------------------------------------------------   [779]
P._nigra_4           --------------------------------------------------   [757]
P._nigra_5           --------------------------------------------------   [781]
P._nigra_2           --------------------------------------------------   [779]
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P._angustifolia      ------------------------------------------------  [640]
P._alba_1            TGCAAAACCAAGCCAATCTCAGTCTTCTTCTGAAGATAGTTCAATTGCAT  [871]
P._alba_2            TGCAAAAC-AAGCCAATCTCAGTCTTCTTCT------------------  [842]
P._alba_3            ------------------------------------------------  [732]
P._tremuloides_1     ------------------------------------------------  [734]
P._grandidentata_3   ------------------------------------------------  [746]
P._grandidentata_4   ------------------------------------------------  [670]
P._davidiana_2       ------------------------------------------------  [661]
P._davidiana_1       ------------------------------------------------  [638]
P._davidiana_5       ------------------------------------------------  [606]
P._tremuloides_2     ------------------------------------------------  [636]
P._tremula_1         ------------------------------------------------  [735]
P._maximowiczii_1    ------------------------------------------------  [542]
P._balsamifera_2     ------------------------------------------------  [513]
P._laurifolia_2      ------------------------------------------------  [747]
P._laurifolia_1      ------------------------------------------------  [777]
P._trichocarpa_1     TGCAAAACCAAGTCAATCTCAGTCTTCTT--------------------  [848]
P._trichocarpa_2     ------------------------------------------------  [745]
P._szechuanica       ------------------------------------------------  [746]
P._deltoides_1       TGCAAAACCAAGCCAATCTCAGTCTTCTTCT------------------  [835]
P._sargentii         TGCAAAACCAAGCCAATCTCAGTCTTCTTCTGAAGATAGTTCAATTGCAT  [869]
P._deltoides_2       ------------------------------------------------  [746]
P._balsamifera_1     TGCAAAACCAAGCCAATCTCAGTCTTCTTCTGAAGATAGTTCAATTGCAT  [856]
S._integra           TGCACAACCAAGTCAATCTCAGTCTTCTTCTGAAGATAGTTCAATTGCAT  [862]
S._nigra             ------------------------------------------------  [510]

[                          ]
[                          ]

P._cathayana         -------   [620]
P._simonii           -------   [851]
P._nigra_1           -------   [814]
P._nigra_3           -------   [779]
P._nigra_4           -------   [757]
P._nigra_5           -------   [781]
P._nigra_2           -------   [779]
P._angustifolia      -------   [640]
P._alba_1            CAAGCTT   [878]
P._alba_2            -------   [842]
P._alba_3            -------   [732]
P._tremuloides_1     -------   [734]
P._grandidentata_3   -------   [746]
P._grandidentata_4   -------   [670]
P._davidiana_2       -------   [661]
P._davidiana_1       -------   [638]
P._davidiana_5       -------   [606]
P._tremuloides_2     -------   [636]
P._tremula_1         -------   [735]
P._maximowiczii_1    -------   [542]
P._balsamifera_2     -------   [513]
P._laurifolia_2      -------   [747]
P._laurifolia_1      -------   [777]
P._trichocarpa_1     -------   [848]
P._trichocarpa_2     -------   [745]
P._szechuanica       -------   [746]
P._deltoides_1       -------   [835]
P._sargentii         CAAGCTT   [876]
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P._deltoides_2          -------    [746]
P._balsamifera_1        CAAGCTT    [863]
S._integra              CAAGCTT    [869]
S._nigra                -------    [510]
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Appendix 2. 6 Nucleotide sequence data matrix of SCISSR fragment PIS9A6.

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[                          10         20         30         40         50]
[                          .          .          .          .          .]

P._nigra_1                ACCAGCTTAGCTTTC-AATTCCTTCAAGCTCAAGGCTTCTC-AAACAAGC    [48]
P._nigra_5                ------------------TTCCTTCA-GCTCA-GGCTTCTCAAAACAAGC    [30]
P._nigra_4                --------AGCTTTC-AATTCCTTCAAGCTCAAGGCTTCTCAAAACAAGC    [41]
P._nigra_2                --CAGCTTAGCTTTCC-ATTCCTTCAAGCTCA-GGCTTCTCAAAACAAGC    [46]
P._alba_1                 -CCAGCTTAGCTTTCC-ATTCCTTCA-GCTCAACGCTTCTCAAAACAAGC    [47]
P._tremuloides_2          GCCAGCTTAGCTTTCC-ATTCCTTCAAGCTCA-CGCTTCT-AAAACAAGC    [47]
P._alba_3                 -------------------------AGCTCAACGCTTCTCAAAACAAGC    [24]
P._grandidentata_2        GCCAGCTTAGCTTTCCAATTCCTTCAAGCTCAACGCTTCTCAAAACAAGC    [50]
P._grandidentata_4        GCCACCTTAGCTTTC-AATTCCTTCAAGCTCAACGCTTCTCAAAACAAGC    [49]
P._alba_2                 ---------------------------------CGCTTCTCAAAACAAGC    [17]
P._davidiana_3            ---CGCTTAGCTTTCC-ATTCCTTCAAGCTCAACGCTTCTCAAAACAAGC    [46]
P._tremuloides_1          GCCAGCTTAGCTTTCCAATTCCTTCAAGCTCAACGCTTCTCAAAACAAGC    [50]
P._grandidentata_3        GCCANCTTAGCTTTCCAATTCCTTCAAGCTCAACGCTTCTCAAAACAAGC    [50]
P._tremula_1              GCCAGCTTAGCTTTCCAATTCCTTCAAGCTCAACGCTTCTCAAAACAAGC    [50]
P._laurifolia_1           ACCAGCTTAGCTTTCC-ATTCCTTCAAGCTCAACGCTTCTCAAAAC----    [45]
P._tristis                -CCAGCTTAGCTTTC-AATTCCTTCAAGCTCA-CGCTTCTCAAAACAAGC    [47]
P._balsamifera_2          -CCAGCTTAGCTTTC-AATTCCTTC-AGCTCAACGCTTCTC-AAACAAGC    [46]
P._balsamifera_1          -CCAGCTTAGCTTTC-AATTCCTTC-AGCTCAACGCTTCTCAAAACAAGC    [47]
P._cathayana              ACCAGCTTAGCTTTCC-ATTCCTTC-AGCTCA-CGCTTCTCAAAACAAGC    [47]
P._nigra_3                --------------------------------------------------    [0]
S._integra                -CCAGCTTAGCTT----------------------CTTC----AACTAGA    [23]
P._laurifolia_2           ---CGCTTAGCTTTCC-ATTCCTTCAAGCTCA-CGCTTCT-AAAAC-T-C    [42]
P._balsamifera_3          ACCAGCTTAGCTTTCC-ATTCCTTC-AGCTCA-CGCTTCTC-AAACAAGC    [46]
P._davidiana_4            ------------------------------------TCTCAAAACAAGC    [13]
P._trichocarpa_2          -----------TTTCC-ATTCCTTC-AGCTCAACGCTTCTC-AAACATGC    [36]
P._szechuanica            --CAGCTTAGCTTTC-AATTCCTTCAAGCTCAACGCTTCTCAAAACATGC    [47]
P._angustifolia           ----------CTTTCC-ATTCCTTCAAGCTCA-CGCTTCTC-AAACAAGC    [37]
P._trichocarpa_1          ACCAGCTTAGCTTTCC-ATTCCTTCAAGCTCAACGCTTCTCAAAACAAGC    [49]
P._maximowiczii_1         ----GCTTAGCTTTCC-ATTCCTTCAAGCTCAACGCTTCTC-AAACAAGC    [44]
P._maximowiczii_2         -----CTTAGCTTTCC-ATTCCTTCAAGCTCAACGCTTCTC-AAACAAGC    [43]
P._fremontii              -CCAGCTTAGCTTTCCAATTCCTTCAAGCTCAACGCTTCTCAAAACAAGC    [49]
P._angulata               -CCAGCTTAGCTTTC-AATTCCTTCA-GCTCAACGCTTCTCAAAACAAGC    [47]
P._deltoides_2            -----------TTTCC-ATTCCTTCAAGCTCAACGCTTCTC-AAACAAGC    [37]
P._deltoides_1            ACCAGCTTAGCTTTCC-ATTCCTTC-AGCTCA-CGCTTCTCAAAACAAGC    [47]
P._sargentii              -CCAGCTTAGCTTTCCAATTCCTTCAAGCTCA-CGCTTCTCAAAACAAGC    [48]

[                          60         70         80         90        100]
[                          .          .          .          .          .]

P._nigra_1                TCTCAGATCTCTCTCGACTGGACCCCGTTTCTCTTTTACCCAAAAAAA--    [96]
P._nigra_5                TCTCAGATCTCTCTCGACTGGACCCCGTTTCTCTTTTACCCAAAAAAA--    [78]
P._nigra_4                TCTCAGATCTCTCTCGACTGGACCCCGTTTCTCTTTTACCCAAAAAAA--    [89]
P._nigra_2                TCTCAGATCTCTCTCGACTGGACCCCGTTTCTCTTTTACCCAAAAAAA--    [94]
P._alba_1                 TCTCAGATCTCTCTCGACTGGACCTCATTTCTCTTTTACCCAAAAAAA--    [95]
P._tremuloides_2          TCTCAGATCTCTCTCGACTGGACCTCATTTCTCTTTTACCCAAAAAAA--    [95]
P._alba_3                 TCTCAGATCTCTCTCGACTGGACCTCATTTCTCTTTTACCCAAAAAAA--    [72]
P._grandidentata_2        TCTCAGATCTCTCTCGACTGNACCCCATTTCTCTTTTACCCAAAAAAA--    [98]
P._grandidentata_4        TCTCAGATCTCTCTCGACTGAACCCCATTTCTCTTTTACCCAAAAAAA--    [97]
P._alba_2                 TCTCAGATCTCTCTCGACTGGACCTCATTTCTCTTTTACCCAAAAAAA--    [65]
P._davidiana_3            TCTCAGATCTCTCTCGACTGGACCCCATTTCTCTTTTACCCAAAAAAA--    [94]
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P._tremuloides_1      TCTCAGATCTCTCTCGACTGGACCNCATTTCTCTTTTACCCAAAAAAA--   [98]
P._grandidentata_3    TCTCAGATCTCTCTCGACTGNACCCCATTTCTCTTTTACCCAAAAAAA--   [98]
P._tremula_1          TCTCAGATCTCTCTCGACTGGACCCCATTTCTCTTCTACCCAAAAAAA--   [98]
P._laurifolia_1       TCTCAGATCTCTCTCGACTGGACCCCATTTCTCTTTTACCCAAAAAAA--   [93]
P._tristis            TCTCAGATCTCTCTCGACTGGACCCCATTTCTCTTTTACCCAAAAAAA--   [95]
P._balsamifera_2      TCTCAGATCTCTCTCGACTGGACCCCATTTCTCTTTTACCCAAAAAAA--   [94]
P._balsamifera_1      TCTCAGATCTCTCTCGACTGGACCCCATTTCTCTTTTACCCAAAAAAA--   [95]
P._cathayana          TCTCAGATCTCTCTCGACTGGACCCCATTTCTCTTTTACCCAAAAAAA--   [95]
P._nigra_3            ---------TCTCTCGACTGGACCCCGTTTCTCTTTTACCCAAAAAAA--   [39]
S._integra            TCATTGATCTCTCTCGAGTGGACCCCATTTCTCTTTTACCTTACAAAA--   [71]
P._laurifolia_2       TCA--GATCTCTCTCGACTGGACCCCATTTCTCTTTTACCCAAAAAAA--   [88]
P._balsamifera_3      TCTCAGATCTCTCTCGACTGGACCCCATTTCTCTTTTACCCAAAAAAA--   [94]
P._davidiana_4        TCTCAGATCTCTCTCGACTGGACCCCATTTCTCTTTTACCCAAGAAAA--   [61]
P._trichocarpa_2      TCTCAGATCTCTCTCGACTGGACCCCATTTCTCTTTTACCCAAAAAAA--   [84]
P._szechuanica        TCTCAGATCTCTCTCGACTGGACCCCATTTCTCTTTTACCCAAAAAAA--   [95]
P._angustifolia       TCTCAGATCTCTCTCGACTGGACCCCATTTCTCTTTTACCCAAAAAAA--   [85]
P._trichocarpa_1      TCTCAGATCTCTCTCGACTGGACCCCATTTCTCTTTTACCCAAAAAAA--   [97]
P._maximowiczii_1     TCTCAGATCTCTCTTGACTGGACCCTATTTCTCTTTTACCCAAAAAAA--   [92]
P._maximowiczii_2     TCTCAGATCTCTCTCGACTGGACCCTATTTCTCTTTTACCCAAAAAAA--   [91]
P._fremontii          TCTCAGATCTCTCTCGACTGGACCCCATTTCTCTTTTACCCAAAAAAAA-   [98]
P._angulata           TCTCAGATCTCTCTCGACTGGACCCCATTTCTCTTTTACCCAAAAAAAA-   [96]
P._deltoides_2        TCTCAGATCTCTCTCGACTGGACCCCATTTCTCTTTTACCCAAAAAAAAA  [87]
P._deltoides_1        TCTCAGATCTCTCTCGACTGGACCCCATTTCTCTTTTACCCAAAAAAAAA  [97]
P._sargentii          TCTCAGATCTCTCTCGACTGGACCCCATTTCTCTTTTACCCAAAAAAAAA  [98]

[                              110       120       130       140       150]
[                              .         .         .         .         .]

P._nigra_1            -GCATGGTCGGACCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [145]
P._nigra_5            -GCATGGTCGGACCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [127]
P._nigra_4            -GCATGGTCGGACCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [138]
P._nigra_2            -GCATGGTCGGACCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [143]
P._alba_1             -GCATGGTCGGGCCATCACTCGACACCACCAAGCTCCATCTCAAACCCCC   [144]
P._tremuloides_2      -GCATGGTCGGACCATCACTCGACACCACCAAGCTCCATCTCAAACCCCC   [144]
P._alba_3             -GCATGGTCGGGCCATCACTCGACACCACCAAGCTCCATCTCAAACCCCC   [121]
P._grandidentata_2    -GCATGGTCGGGCCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [147]
P._grandidentata_4    -GCATGGTCGGGCCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [146]
P._alba_2             -GCATGGTCGGGCCATCACTCGACACCACCAAGCTCCATCTCAAACCCCC   [114]
P._davidiana_3        -GCATGGTCGGACCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [143]
P._tremuloides_1      -GCATGGTCGGNCCATCACTNGACACCACCAAGCTCCATCTCAAACCCCC   [147]
P._grandidentata_3    -GCATGGTCGGNCCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [147]
P._tremula_1          -GCATGGTCGGACCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [147]
P._laurifolia_1       -GCATGGTCGGACCATCACTTGACACCTCCAAGCTCCATCTCAAACCCCC   [142]
P._tristis            -GAATGGTCGGACGATCACTTGACACCACCAAGCTCCTTCTCAAACCCCC   [144]
P._balsamifera_2      -GCATGGTCGGACCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [143]
P._balsamifera_1      -GCATGGTCGGACCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [144]
P._cathayana          -GCATGGTCGGACCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [144]
P._nigra_3            -GCATGGTCGGACCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [88]
S._integra            -GCATGGTCGGACCACCACTTGACACCACCAAGCTCCGTCTCAAACCGCC   [120]
P._laurifolia_2       -GCATGGTCGGACCATCACTTGACACCTCCAAGCTCCATCTCAAACCCCC   [137]
P._balsamifera_3      -GCATGGTCGGACCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [143]
P._davidiana_4        -GCATGGTCGGGCCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [110]
P._trichocarpa_2      -GCATGGTCGGACCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [133]
P._szechuanica        -GCATGGTCGGACCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [144]
P._angustifolia       -GCATGGTCGGACCATCACTTGACACCGCCAAGCTCCATCTCAAACCCCC   [134]
P._trichocarpa_1      -GCATGGTCGGACCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [146]
P._maximowiczii_1     -GCATGGTCGGACCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [141]
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P._maximowiczii_2     -GCATGGTCGGACCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [140]
P._fremontii          -GCATGGTCGGACCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [147]
P._angulata           -GCATGGTCGGACCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [145]
P._deltoides_2        AGCATGGTCGGACCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [137]
P._deltoides_1        AGCATGGTCGGACCATCACTTGACACCACCAAGCTCCATCTCAAACCCCC   [147]
P._sargentii          AGCATGGGCGGAACATTACTTGAAACCCCCAAGCTTCATTTTAAAACCCC   [148]

[                              160       170       180       190       200]
[                              .         .         .         .         .]

P._nigra_1            CTCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [195]
P._nigra_5            CTCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [177]
P._nigra_4            CTCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [188]
P._nigra_2            CTCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [193]
P._alba_1             CTCGTCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [194]
P._tremuloides_2      CTCGTCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [194]
P._alba_3             CTCGTCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [171]
P._grandidentata_2    TTCGTCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [197]
P._grandidentata_4    TTCGTCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [196]
P._alba_2             CTCGTCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [164]
P._davidiana_3        CTCGTCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [193]
P._tremuloides_1      CTCGTCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [197]
P._grandidentata_3    TTCGTCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [197]
P._tremula_1          CTCGTCTGCCACCCCCACCATCAAGTTCCTCTGCAGTTACGGTGGCAAGA   [197]
P._laurifolia_1       CTCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [192]
P._tristis            CTCTTCTGCCACCCCCACCATCAAATTCTTCTGCAGTTACGGTGGCAAGA   [194]
P._balsamifera_2      CTCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [193]
P._balsamifera_1      CTCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [194]
P._cathayana          CTCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [194]
P._nigra_3            CTCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [138]
S._integra            CTCATCTGCCACCCCCACCATGAAGTTCCTCTGCAGCTATGGTGGCAAGA   [170]
P._laurifolia_2       CTCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [187]
P._balsamifera_3      CTCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [193]
P._davidiana_4        ATCGTCTGCCACCCCCACCATCAAGTTCCTCTGCAGTTACGGTGGCAAGA   [160]
P._trichocarpa_2      CTCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [183]
P._szechuanica        CTCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [194]
P._angustifolia       TTCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [184]
P._trichocarpa_1      CTCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [196]
P._maximowiczii_1     CTCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [191]
P._maximowiczii_2     CTCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [190]
P._fremontii          TTCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [197]
P._angulata           TTCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [195]
P._deltoides_2        TCCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [187]
P._deltoides_1        TCCATCTGCCACCCCCACCATCAAATTCCTCTGCAGTTACGGTGGCAAGA   [197]
P._sargentii          TCCATTTGGCACCCCCACCATTAAATTCCTTTGGAGTTACGGGGGGAAAA   [198]

[                              210       220       230       240       250]
[                              .         .         .         .         .]

P._nigra_1            TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCAAGGTGGCGAAACC   [245]
P._nigra_5            TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCAAGGTGGCGAAACC   [227]
P._nigra_4            TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCAAGGTGGCGAAACC   [238]
P._nigra_2            TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCAAGGTGGCGAAACC   [243]
P._alba_1             TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCACGGTGGCGAAACC   [244]
P._tremuloides_2      TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCACGGTGGCGAAACC   [244]
P._alba_3             TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCACGGTGGCGAAACC   [221]
P._grandidentata_2    TCATCCCCCGTTTCCCAGATGGCAAACTCCGCTACCACGGTGGCGAAACC   [247]
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P._grandidentata_4    TCATCCCCCGTTTCCCAGATGGCAAACTCCGCTACCACGGTGGCGAAACC   [246]
P._alba_2             TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCACGGTGGCGAAACC   [214]
P._davidiana_3        TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCACGGTGGCGAAACC   [243]
P._tremuloides_1      TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCACGGTGGCGAAACC   [247]
P._grandidentata_3    TCATCCCCCGTTTCCCAGATGGCAAACTCCGNTACCACGGTGGCGAAACC   [247]
P._tremula_1          TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCACGGTGGCGAAACC   [247]
P._laurifolia_1       TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCACGGTGGCGAAACC   [242]
P._tristis            TCATCCCCCGTTTCCCAGATGGCAAACTCCTTTACCACGGTGGCGAAAAA   [244]
P._balsamifera_2      TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCACGGTGGCGAAACC   [243]
P._balsamifera_1      TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCACGGTGGCGAAACC   [244]
P._cathayana          TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCACGGTGGCGAAACC   [244]
P._nigra_3            TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCAAGGTGGCGAAACC   [188]
S._integra            TCATCCCCCGCTTCCCAGATGGAAAACTCCGTTACCTAGGTGGCGAAACC   [220]
P._laurifolia_2       TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCACGGTGGCGAAACC   [237]
P._balsamifera_3      TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCACGGTGGCGAAACC   [243]
P._davidiana_4        TCATCCCCCGTTTCCCAGATGGAAAACTCCGTTACCACGGTGGCGAAACC   [210]
P._trichocarpa_2      TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCACGGTGGCGAAACC   [233]
P._szechuanica        TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCACGGTGGCGAAACC   [244]
P._angustifolia       TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCACGGTGGCGAAACC   [234]
P._trichocarpa_1      TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCACGGTGGCGAAACC   [246]
P._maximowiczii_1     TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCACGGTGGCGAAACC   [241]
P._maximowiczii_2     TCATCCCCCGTTTCCCAGATGGCAAACTCCGTTACCACGGTGGCGAAACC   [240]
P._fremontii          TCATCCCCCGTTTCCCAAATGGCAAACTCCGTTACCACGGTGGCAAAACC   [247]
P._angulata           TCATCCCCCGTTTCCCAAATGGCAAACTCCGTTACCACGGTGGCAAAACC   [245]
P._deltoides_2        TCATCCCCCGTTTCCCAAATGGCAAACTCCGTTACCACGGTGGCAAAACC   [237]
P._deltoides_1        TCATCCCCCGTTTCCCAAATGGCAAACTCCGTTACCACGGTGGCAAAACC   [247]
P._sargentii          ACCTCCCCCGTTTCCCAAAAGGGAAAATCCCTTACCCCGGGGGGAAAACC   [248]

[                              260       270       280       290       300]
[                              .         .         .         .         .]

P._nigra_1            CGTGTCCTCGGTGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTA   [295]
P._nigra_5            CGTGTCCTCGGTGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTA   [277]
P._nigra_4            CGTGTCCTCGGTGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTA   [288]
P._nigra_2            CGTGTCCTCGGTGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTA   [293]
P._alba_1             CGTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTACTG   [294]
P._tremuloides_2      CGTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [294]
P._alba_3             CGTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTACTG   [271]
P._grandidentata_2    CGTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [297]
P._grandidentata_4    CGTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [296]
P._alba_2             CGTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [264]
P._davidiana_3        CGTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [293]
P._tremuloides_1      CGTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [297]
P._grandidentata_3    CGTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [297]
P._tremula_1          CGTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [297]
P._laurifolia_1       CGTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [292]
P._tristis            CGTGTCCTCGGCGTCGAACATTCCATCTCCTTTGCTGGTGAGTCTTATTG   [294]
P._balsamifera_2      CGTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [293]
P._balsamifera_1      CGTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [294]
P._cathayana          CGTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [294]
P._nigra_3            CGTGTCCTCGGTGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTA   [238]
S._integra            CGTGTCCTTGGCGTCGAACGGTCCATCTCCTTTGCAGGTCAATCTTGTTA   [270]
P._laurifolia_2       CGTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [287]
P._balsamifera_3      CGTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [293]
P._davidiana_4        CGTGTCCTCGGCGTCAAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [260]
P._trichocarpa_2      CGTGTCCTCGGCGTCAAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [283]
P._szechuanica        CTTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [294]
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P._angustifolia       CGTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [284]
P._trichocarpa_1      CGTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [296]
P._maximowiczii_1     CGTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [291]
P._maximowiczii_2     CGTGTCCTCGGCGTCGAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTG   [290]
P._fremontii          CGTGTCCTCGGTGTCAAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTA   [297]
P._angulata           CGTGTCCTCGGTGTCAAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTA   [295]
P._deltoides_2        CGTGTCCTCGGTGTCAAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTA   [287]
P._deltoides_1        CGTGTCCTCGGTGTCAAACGTTCCATCTCCTTTGCTGGTCAGTCTTATTA   [297]
P._sargentii          CGGGTCCCCGGGGTCAAAAGTTCCATCCCCTTTGGGGGGCAGTCTTATTA   [298]

[                              310        320        330        340        350]
[                              .          .          .          .          .]

P._nigra_1            TATATCACTA----TTAATTTCCACTATTTTCCCTTTTCTTTTCTTTTAG   [341]
P._nigra_5            TATATCACTA----TTAATTTCCACTATTTTCCCTTTTCTTTTCTTTTAG   [323]
P._nigra_4            TATATCACTA----TTAATTTCCACTATTTTCCCTTTTCTTTTCTTTTAG   [334]
P._nigra_2            TATATCACTA----TTAATTTCCACTATTTTCCCTTTTCTTTTCTTTTAG   [339]
P._alba_1             TATATCACTA----CTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [340]
P._tremuloides_2      TATATCACTA----CTAAATTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [340]
P._alba_3             TATATCACTA----CTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [317]
P._grandidentata_2    TATATCACTA----CTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [343]
P._grandidentata_4    TATATCACTA----CTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [342]
P._alba_2             TATATCACTA----CTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [310]
P._davidiana_3        TATATCACTA----ATAATTTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [339]
P._tremuloides_1      TATATCACTA----CTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [343]
P._grandidentata_3    TATATCACTA----CTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [343]
P._tremula_1          TATATCACTA----CTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [343]
P._laurifolia_1       TATATCACTA----CTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [338]
P._tristis            TATATCACTA----CTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTCG   [340]
P._balsamifera_2      TATATCACTA----CTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTCG   [339]
P._balsamifera_1      TATATCACTA----CTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTCG   [340]
P._cathayana          TATATCACTA----CTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTCG   [340]
P._nigra_3            TATATCACTA----TTAATTTCCACTATTTTCCCTTTTCTTTTCTTTTAG   [284]
S._integra            TATATCACTAGCTATTAATTTCTACTATTTCCCCTTTTCTTTTCTTTTTG   [320]
P._laurifolia_2       TATATCACTA----CTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [333]
P._balsamifera_3      TATATCACTA----CTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [339]
P._davidiana_4        TATATCACTA----CTAATTTCC---------CCTTTTCTTTTCTTTTTG   [297]
P._trichocarpa_2      TATATCACTA----CTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [329]
P._szechuanica        TATATCACTA----CTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [340]
P._angustifolia       TATATCACTA----TTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [330]
P._trichocarpa_1      TATATCACTA----CTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [342]
P._maximowiczii_1     TATATCACTA----CTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTAG   [337]
P._maximowiczii_2     TATATCACTA----CTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTAG   [336]
P._fremontii          TATATCACTA----TTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [343]
P._angulata           TATATCACTA----TTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [341]
P._deltoides_2        TATATCACTA----TTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [333]
P._deltoides_1        TATATCACTA----TTAATTTCCACTATTTCCCCTTTTCTTTTCTTTTTG   [343]
P._sargentii          AAAATCACTA----TTAATTTCCCCTATTTCCCCTTTTCTTTTCTTTTTG   [344]

[                              360        370        380        390        400]
[                              .          .          .          .          .]

P._nigra_1            CCAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTCGTTGTGTGTGAAT   [391]
P._nigra_5            CCAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTCGTTGTGTGTGAAT   [373]
P._nigra_4            CCAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTCGTTGTGTGTGAAT   [384]
P._nigra_2            CCAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTCGTTGTGTGTGAAT   [389]
P._alba_1             CTAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTATTTGTGTGTGAAT   [390]
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P._tremuloides_2      CTAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTATTTGTGTGTGAAT   [390]
P._alba_3             CTAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTATTTGTGTGTGAAT   [367]
P._grandidentata_2    CTAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTATTTGTGTGTGAAT   [393]
P._grandidentata_4    CTAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTATTTGTGTGTGAAT   [392]
P._alba_2             CTAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTATTTGTGTGTGAAT   [360]
P._davidiana_3        CTAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTATTTGTGTGTGAAT   [389]
P._tremuloides_1      CTAATTCCTTCTTTAGCTTTTACCGTATCAANTGTTATTTGTGTGTGAAT   [393]
P._grandidentata_3    CTAATTCCTTCTTTAGCTTTTACCGTATCAANTGTTATTTGTGTGTGAAT   [393]
P._tremula_1          CTAATTCCTTCTTTAGCTTTTACCGNATCAACTGTTATTTGTGTGTGAAT   [393]
P._laurifolia_1       CTAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTATTTTTGTGTGAAT   [388]
P._tristis            CTAATTCCTTCTTTAGCTTTTACCATATCAAGTGTTATTTGTGTGTGAAT   [390]
P._balsamifera_2      CTAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTATTTGTGTGTGAAT   [389]
P._balsamifera_1      CTAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTATTTGTGTGTGAAT   [390]
P._cathayana          CTAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTATTTGTGTGTGAAT   [390]
P._nigra_3            CCAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTCGTTGTGTGTGAAT   [334]
S._integra            CCAATTCCTTCTTTAGCTTTTACCGAATCGAGTATAATTTCTGTGTGAAT   [370]
P._laurifolia_2       CTAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTATTTTTGTGTGAAT   [383]
P._balsamifera_3      CTAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTATTTGTGTGTGAAT   [389]
P._davidiana_4        CTAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTATTTGTGTGTGAAT   [347]
P._trichocarpa_2      CTAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTATTTGTGTGTGAAT   [379]
P._szechuanica        CTAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTATTTGTGTGTGAAT   [390]
P._angustifolia       CTAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTATTTGTGTGTGAAT   [380]
P._trichocarpa_1      CTAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTATTTGTGTGTGAAT   [392]
P._maximowiczii_1     CTAATTCCTTCTTTAGCTTTTACCGTGTCAAGTGTTATTTGTGTGTGAAT   [387]
P._maximowiczii_2     CTAATTCCTTCTTTAGCTTTTACCGTGTCAAGTGTTATTTGTGTGTGAAT   [386]
P._fremontii          CTAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTATTTGTGTGTGAAT   [393]
P._angulata           CTAATTCCTTCTTTAGCTTTTACCGTATCAAGTGTTATTTGTGTGTGAAT   [391]
P._deltoides_2        CTAATTCCTTCTTTAGCTTTTACCGTACCAAGTGTTATTTGTGTGTGAAT   [383]
P._deltoides_1        CTAATTCCTTCTTTAGCTTTTACCGTACCAAGTGTTATTTGTGTGTGAAT   [393]
P._sargentii          CCAATTCCTTCTTTAACTTTTACCGTAACAAGGGTTATTTGGGGGGGAAA   [394]

[                              410        420        430        440        450]
[                              .          .          .          .          .]

P._nigra_1            ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCTTGTTTGC   [433]
P._nigra_5            ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCTTGTTTGC   [415]
P._nigra_4            ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCTTGTTTGC   [426]
P._nigra_2            ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCTTGTTTGC   [431]
P._alba_1             AT-----------------------------------TTTTGCGTGTGTGC   [406]
P._tremuloides_2      AT-----------------------------------TTTTGCGTGTGTGC   [406]
P._alba_3             AT-----------------------------------TTTTGCGTGTGTGC   [383]
P._grandidentata_2    AT-----------------------------------TTTTGCGTGTGTGC   [409]
P._grandidentata_4    AT-----------------------------------TTTTGCGTGTGTGC   [408]
P._alba_2             AT-----------------------------------TTTTGCGTGTGTGC   [376]
P._davidiana_3        ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCGTGTGTGC   [431]
P._tremuloides_1      AT-------------------------------------------------   [395]
P._grandidentata_3    -------------------------------------------------   [393]
P._tremula_1          ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCGTGTGTGC   [435]
P._laurifolia_1       ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCGTGTGTGC   [430]
P._tristis            ATG--------GAGTAAGTGATACGGGGGGGAATATTTTTGCGTGTGTGC   [432]
P._balsamifera_2      ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCGTGTGTGC   [431]
P._balsamifera_1      ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCGTGTGTGC   [432]
P._cathayana          ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCGTGTGTGC   [432]
P._nigra_3            ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCTTGTTTGC   [376]
S._integra            ATGTGATGATGGAGTAAGTGATATGCGGTGGAATATTTTTGCGTGTGTGC   [420]
P._laurifolia_2       ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCGTGTGTGC   [425]
P._balsamifera_3      ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCTTGTGTGC   [431]
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P._davidiana_4          ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCGTGTGTGC   [389]
P._trichocarpa_2        ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCGTGTGTGC   [421]
P._szechuanica          ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCTTGTGTGC   [432]
P._angustifolia         ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCTTGTGTGC   [422]
P._trichocarpa_1        ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCTTGTGTGC   [434]
P._maximowiczii_1       ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCTTGTGTGC   [429]
P._maximowiczii_2       ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCTTGTGTGC   [428]
P._fremontii            ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCTTTTGTGC   [435]
P._angulata             ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCTTTTGTGC   [433]
P._deltoides_2          ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCTTGTGTGC   [425]
P._deltoides_1          ATG--------GAGTAAGTGATACGGGGTGGAATATTTTTGCTTGTGTGC   [435]
P._sargentii            ATG--------GAATAAGTGAAACGGGGGGGAAAATTTTTGCTTGGGGGC   [436]

[                                460        470        480        490        500]
[                                .          .          .          .          .]

P._nigra_1              TACACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [483]
P._nigra_5              TACACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [465]
P._nigra_4              TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [476]
P._nigra_2              TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [481]
P._alba_1               TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [456]
P._tremuloides_2        TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [456]
P._alba_3               TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [433]
P._grandidentata_2      TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [459]
P._grandidentata_4      TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [458]
P._alba_2               TATACAGAGTTATTGTTGAAGCTTGGAGGGTTACGTGGGACATCGGTGAA   [426]
P._davidiana_3          TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [481]
P._tremuloides_1        --------------------------------------------------   [395]
P._grandidentata_3      --------------------------------------------------   [393]
P._tremula_1            TATACAGAGTTATTGTNGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [485]
P._laurifolia_1         TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [480]
P._tristis              TATACAAAGTTATTGTTGAAGCTTGTAGAGTTACTTGGGACATCAGTGAA   [482]
P._balsamifera_2        TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [481]
P._balsamifera_1        TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [482]
P._cathayana            TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [482]
P._nigra_3              TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [426]
S._integra              TATACAGAGTTATCGTTGAAGCTCGGTGAGTTATGTGGGACATCGGCGAG   [470]
P._laurifolia_2         TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [475]
P._balsamifera_3        TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [481]
P._davidiana_4          TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [439]
P._trichocarpa_2        TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [471]
P._szechuanica          TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [482]
P._angustifolia         TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [472]
P._trichocarpa_1        TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [484]
P._maximowiczii_1       TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [479]
P._maximowiczii_2       TATACAGAGTTATTGTTGAAGCTTGGAGAGTTACGTGGGACATCGGTGAA   [478]
P._fremontii            TATACAAAGTTATTGTTGAAGCTTGAAAAGTTACGTGGGACATCGGTGAA   [485]
P._angulata             TATACAAAGTTATTGTTGAAGCTTGGAAAGTTACGTGGGACATCGGTGAA   [483]
P._deltoides_2          TATACAAATTTATTGTTGAAGCTTGGAAAGTTACGTGGGACATCGGTGAA   [475]
P._deltoides_1          TATACAAATTTATTGTTGAAGCTTGGAAAGTTACGTGGGACATCGGTGAA   [485]
P._sargentii            CAAAAAAATTTATTGTTTAAACTTGGAAAATTACCTGGGAAATCCGTGAA   [486]

[                                510        520        530        540        550]
[                                .          .          .          .          .]

P._nigra_1              TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGATGCTCTGGTATCGATCA   [533]
P._nigra_5              TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGATGCTCTGGTATCGATCA   [515]
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P._nigra_4              TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGATGCTCTGGTATCGATCA   [526]
P._nigra_2              TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGATGCTCTGGTATCGATCA   [531]
P._alba_1               TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGTATCGATCA   [506]
P._tremuloides_2        TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGTATCGATCA   [506]
P._alba_3               TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGTATCGATCA   [483]
P._grandidentata_2      TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGTATCGATCA   [509]
P._grandidentata_4      TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGTATCGATCA   [508]
P._alba_2               TCTCCGTTGTCAGTTACCAAAAGAAGATCTAGACGCTCTGGTATCGATCA   [476]
P._davidiana_3          TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGTATCGATCA   [531]
P._tremuloides_1        ----------------------------------------------------  [395]
P._grandidentata_3      ----------------------------------------------------  [393]
P._tremula_1            TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGNATCGATCA   [535]
P._laurifolia_1         TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGTATCGATCA   [530]
P._tristis              TATCCTTTGTCATTTGCCAAAAAAAGATCAAGACGCTCTGGTATCAATCA   [532]
P._balsamifera_2        TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGTATCGATCA   [531]
P._balsamifera_1        TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGTATCGATCA   [532]
P._cathayana            TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGTATCGATCA   [532]
P._nigra_3              TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGATGCTCTGGTATCGATCA   [476]
S._integra              TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGTTTCGATCA   [520]
P._laurifolia_2         TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGTATCGATCA   [525]
P._balsamifera_3        TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGTATCGATCA   [531]
P._davidiana_4          TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGTATCGATCA   [489]
P._trichocarpa_2        TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGTATCGATCA   [521]
P._szechuanica          TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGTATCGATCA   [532]
P._angustifolia         TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGTATCGATCA   [522]
P._trichocarpa_1        TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGTATCGATCA   [534]
P._maximowiczii_1       TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGTATCGATCA   [529]
P._maximowiczii_2       TCTCCGTTGTCAGTTGCCAAAAGAAGATCTAGACGCTCTGGTATCGATCA   [528]
P._fremontii            TCTCCGTTGTCAGTTGCCAAAAAAAAATCTAAATGCTCTGGTATCGATCA   [535]
P._angulata             TCTCCGTTGTCAGTTGCCAAAAAAAAATCTAAATGCTCTGGTATCAATCA   [533]
P._deltoides_2          TCTCCGGTGTCAGTTGCCAAAAAAAAATCTAGACGCTCTGGTATCGATCA   [525]
P._deltoides_1          TCTCCGGTGTCAGTTGCCAAAAAAAAATCTAGACGCTCTGGTATCGATCA   [535]
P._sargentii            TCCCCGGGGTCAGTTGCCAAAAAAAAAACTAAACCCCTTGGTATCCATCA   [536]

[                                560       570       580       590       600]
[                                .         .         .         .         .]

P._nigra_1              CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT   [583]
P._nigra_5              CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT   [565]
P._nigra_4              CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT   [576]
P._nigra_2              CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT   [581]
P._alba_1               CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT   [556]
P._tremuloides_2        CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT   [556]
P._alba_3               CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT   [533]
P._grandidentata_2      CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT   [559]
P._grandidentata_4      CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT   [558]
P._alba_2               CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT   [526]
P._davidiana_3          CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT   [581]
P._tremuloides_1        ----------------------------------------------------  [395]
P._grandidentata_3      ----------------------------------------------------  [393]
P._tremula_1            CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT   [585]
P._laurifolia_1         CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT   [580]
P._tristis              CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACTATCAAGCAGCT   [582]
P._balsamifera_2        CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT   [581]
P._balsamifera_1        CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT   [582]
P._cathayana            CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT   [582]
P._nigra_3              CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT   [526]
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S._integra              CCTCCGATGAGGATCTAGCTAATCTCATCGAGGAATACGATCAGGCAGCT    [570]
P._laurifolia_2         CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT    [575]
P._balsamifera_3        CCTCTGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT    [581]
P._davidiana_4          CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT    [539]
P._trichocarpa_2        CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT    [571]
P._szechuanica          CCTCTGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT    [582]
P._angustifolia         CCTCTGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT    [572]
P._trichocarpa_1        CCTCTGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT    [584]
P._maximowiczii_1       CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT    [579]
P._maximowiczii_2       CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACGATCGAGCAGCT    [578]
P._fremontii            CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACAACCGAGCAGCT    [585]
P._angulata             CCTCCGGTGAGGATCTAGCTAATCTCATCAAGGAATACAACCGAGCAGCT    [583]
P._deltoides_2          CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACAATCGAGCAGCT    [575]
P._deltoides_1          CCTCCGGTGAGGATCTAGCTAATCTCATCGAGGAATACAATCGAGCAGCT    [585]
P._sargentii            CCCCCGGGGA----------------------------------------    [546]

[                              610       620       630       ]
[                              .         .         .         ]

P._nigra_1              GGAGCAGCAACACCAACTGCATCTTTAAAGATCAGG---    [619]
P._nigra_5              GGAGCAGCAACACCAACTGCATCTTTAAAGATCAGGGC-    [603]
P._nigra_4              GGAGCAGCAACACCAACTGCATCTTTAAAGATCA-----    [610]
P._nigra_2              GGAGCAGCAACACCAACTGCATCTTTAAAGATCA-----    [615]
P._alba_1               GGAGCAGCAACACCAACTGCATCTTTAAAGATCAG----    [591]
P._tremuloides_2        GGAGCAGCAACACCAACTGCATCTTTAAAGATCAGGGCT    [595]
P._alba_3               GGAGCAGCAACACCAACTGCATCTTTAAAGATCAGGGC-    [571]
P._grandidentata_2      GGAGCAGCAACACCAACTGCATCTTTAAAGATCAGGGC-    [597]
P._grandidentata_4      GGAGCAGCAACACCAACTGCATCTTTAAAGATCAGGGCT    [597]
P._alba_2               GGAGCAGCAACACCAACTGCATCTTTAAAGATCAG----    [561]
P._davidiana_3          GGAGCAGCAACACCAACTGCATCTTTAAAGATCAG----    [616]
P._tremuloides_1        ----------------------------------------    [395]
P._grandidentata_3      ----------------------------------------    [393]
P._tremula_1            GGAGCAGCAACACCAACT----------------------    [603]
P._laurifolia_1         GGAGCAGCAACACCAACTGCATCTTTAAAGAT-------    [612]
P._tristis              GGAGCAGCAACACCAACTGCTTCTTTAA-----------    [610]
P._balsamifera_2        GGAGCAGCAACACCAACTGCATCTTTAAAGATCAGGGC-    [619]
P._balsamifera_1        GGAGCAGCAACACCAACTGCATCTTTAAAGATCA-----    [616]
P._cathayana            GGAGCAGCAACACCAACTGCATCTTTAAAGATCAGG---    [618]
P._nigra_3              GGAGCAGCAACACCAACTGCATCTTTAAAGATCAGGGCT    [565]
S._integra              GGGCAAGCAACACCAACTGCATCTTTAAAGATCAGGGC-    [608]
P._laurifolia_2         GGAGCAGCAACACCAACTGCATCTTTAAAGATCA-----    [609]
P._balsamifera_3        GGAGCAGCAACACCAACTGCATCTTTAAAGATCAGG---    [617]
P._davidiana_4          GGAGCAGCAACACCAACTGCATCTTTAAAGATCAGGGCT    [578]
P._trichocarpa_2        GGAGCAGCAACACCAACTGCATCTTTAAAGATCAGGGC-    [609]
P._szechuanica          GGAGCAGCAACACCAACTGCATCTTTAAAGATCAGGGCT    [621]
P._angustifolia         GGAGCAGCAACACCAACTGCATCTTTAAAGATCAGGGC-    [610]
P._trichocarpa_1        GGAGCAGCAACACCAACTGCATCTTTAAAGATCAGGGCT    [623]
P._maximowiczii_1       GGAGCAGCAACACCAACTGCATCTTTAAAGATCAGG---    [615]
P._maximowiczii_2       GGAGCAGCAACACCAACTGCATCTTTAAAGATCAG----    [613]
P._fremontii            GGAGCAGCAACACCAACTGCATCTTTAAAGATCAG----    [620]
P._angulata             GGAGCAGCAACACCAACTGCATCTTTAAAGATCAGGGCT    [622]
P._deltoides_2          GGAGCAGCAACACCAACTGCATCTTTAAAGATCAG----    [610]
P._deltoides_1          GGAGCAGCAACACCAACTGCATCTTTAAAGATCAGG---    [621]
P._sargentii            ----------------------------------------    [546]
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CHAPTER 3; ASYMMETRICAL NATURAL HYBRIDIZATION BETWEEN *POPULUS DELTOIDES* AND *P. BALSAMIFERA* (SALICACEAE)

ABSTRACT

Natural hybridization has long been recognized as a means for gene flow between species and has important evolutionary consequences. Although hybridization is generally considered to be symmetrical, with both hybridizing species being equally likely to be the male or female parent, several studies have demonstrated the presence of asymmetrical hybridization and introgression from one species to the other. We investigated the direction of natural hybridization between two sympatric forest tree species in North America (*Populus deltoides* and *P. balsamifera*) using species-specific single nucleotide polymorphism (SNP) markers in both the nuclear and chloroplast genomes. All natural hybrid individuals, identified based on morphological traits, had nuclear alleles corresponding to both parental species, while the chloroplast genotypes showed similarity to *P. deltoides*, indicating asymmetrical hybridization with *P. deltoides* as the maternal and *P. balsamifera* as the paternal donor species. This observed asymmetrical hybridization may be attributable to cytonuclear interactions.

Keywords: *Populus*, natural hybridization, asymmetry, SNP markers.

3. 1 INTRODUCTION

Natural hybridization among species influences the evolution of biological diversity in a variety of ways including an increase in intraspecific genetic diversity (Anderson 1948), altering genetic adaptations (Anderson 1948; Stebbins 1950), the origin of new ecotypes or species (Grant 1981; Whitham et al. 1994), and the reinforcement or breakdown of reproductive barriers (Ellstrand and Elam 1993; Rieseberg and Gerber 1995; Levin et al. 1996). Introgression or the transfer of genetic material from one species into another, has been documented in a wide variety of plant and animal taxa (Arnold 1992; Rieseberg and Wendel 1993) and may serve as a source of genetic variation for adaptive evolution (Lewontin and Birch 1966; Grant and Grant 1994, 1996; Wang et al. 1997). Hybridization between species has traditionally been examined for its role in finalising the speciation process through reinforcement of reproductive barriers (Dobzhansky 1940; Mayr 1963; Howard 1993). The evolution of barriers to inter-species gene flow plays significant roles in the process of speciation. Therefore, understanding the patterns of reproductive isolation between species is crucial for gaining insights into the mechanisms that control interspecific gene exchange or natural hybridization.

Inter-species hybridization is one of the common features among many members of the genus *Populus* L, a group of widely distributed forest trees in the northern hemisphere (Barnes 1961; Brayshaw 1965; Eckenwalder 1982, 1996; Barnes and Pregitzer 1985; Stettler et al. 1996 b; Whitham et al. 1996). In North America, several species of the genus *Populus*, particularly species of the sections *Tacamahaca* Spach. and *Aigeiros* Duby, are broadly sympatric (Eckenwalder 1984 a, b), and known to hybridize

extensively (Brayshaw 1965; Ronald et al. 1973 a, b; Eckenwalder 1984 a, b; Rood et al. 1986; Greenaway et al. 1991; Floate 2004).

Through multivariate analysis of leaf morphology of *Populus* species, Rood et al. (1986) showed continuous variation of characters and suggested the hybridization between *P. deltoides*, *P. balsamifera* and *P. angustifolia*. These observations were further confirmed with gas chromatography-mass spectrometry data (Greenaway et al. 1991). Comprehensive analysis of leaf morphological characters of *P. balsamifera*, *P. angustifolia* and *P. deltoides* in the hybrid zones in the riparian forests in southern Alberta suggested bidirectional introgression between *P. angustifolia* and *P. balsamifera*, and unidirectional introgression between *P. balsamifera* and *P. deltoides* (Floate 2004). Molecular data has revealed a similar unidirectional introgression between *P. fremontii* and *P. angustifolia* in the Weber River drainage system in northern Utah (Martinsen et al. 2001; Keim et al. 1989). Although the occurnce of natural hybridization and directionality of the introgression (unidirectional) between *P. deltoides* and *P. balsamifera* has been successfully shown using morphological data, the maternal and paternal parental source of the F1 natural hybrids remains unknown.

In this study, we used both nuclear- and chloroplast-based single nucleotide polymorphisms (SNPs), specific for *P. deltoides* and *P. balsamifera*, to test the hybrid status of morphologicaly identified hybrids, and determine the maternal and paternal parental species of the F_1 hybrids between these two species (known as *P.* x *jackii*) collected from natural stands, to infer the parentage of hybrids in natural populations.

3. 2 MATERIALS AND METHODS

3. 2. 1 Plant material and DNA extraction

Fresh leaf samples were collected from morphologically distinct *P. deltoides*, *P. balsamifera*, first generation progeny (F_1) of controlled-crosses between *P. deltoides* and *P. balsamifera*, and *P.* x *jackii* occurring in natural stands (Tables 3. 1 and 3. 2). These hybrids are usually recognised by their broadly ovate leaves with heart-shaped bases. In this study, we used natural hybrids that can be morphologically distinguished from the parental species. Thus, complex backcross hybrids that are morphologically indistinguishable from the parental species may not have been sampled for the present study. However, the exclusion of such hybrids in our samples was not expected to affect the interpretation of our results. The collected leaf samples were stored at –80 ℃ prior to extraction of DNA. The total genomic DNA was extracted from frozen leaf tissue using the methods of Doyle and Doyle (1987) and Dayanandan et al. (1997).

3. 2. 2 Selection and validation of SNPs

Molecular markers represent a powerful tool for identifying hybrid taxa (Rieseberg and Wendel 1993). Species-specific molecular markers derived from both biparentally-inherited nuclear and uniparentally-inherited organelles such as the chloroplast, which is maternally inherited in *Populus* (Rajora and Danick 1995), are invaluable for detecting the parentage of putative hybrids. In particular, single nucleotide polymorphisms (SNPs) or DNA sequence variations that occur at a specific site in the genome (Cho et al. 1999; Griffin and Smith 2000) are useful for detecting the hybrids.

Based on nucleotide sequence matrices of two noncoding regions of the chloroplast DNA (intron of *trnL* and intergenic region of *trnT-trnL*) and *ITS2* of the nuclear rDNA from a previous study of *Populus* (Hamzeh and Dayanandan 2004), four chloroplast and two nuclear SNPs specific to *P. balsamifera* and *P. deltoides* were identified (Table 3. 3). The nrSNP1 and nrSNP2 were located within the ITS2 region of the nuclear rDNA. The cpSNP1 was located within the intergenic region of *trnT-trnL*, and cpSNP2, cpSNP3 and cpSNP4 were located in the *trnL* intron of the chloroplast genome. The selected nuclear and chloroplast SNPs were further characterized for use as reliable molecular markers by genotyping 30 individuals of *P. deltoides* and 10 individuals of *P. balsamifera*. The inheritance pattern of SNPs was determined by genotyping 20 individuals selected from progenies of controlled crosses between *P. deltoides* and *P. balsamifera* and their parents.

3. 2. 3 Template preparation

Target genomic regions containing SNPs were amplified by the polymerase chain reaction (PCR). The oligonucleotide primers a, c, and d (Taberlet et al. 1991) and *trn*CR (Hamzeh and Dayanandan 2004) for the chloroplast genome and primers *ITS3* (Becerra and Venable 1999) and *ITS 28kj* modified from Culling (1992) for the *ITS2* region were used for PCR amplification. Amplification reactions contained 230 μM dNTP, 2.5 mM $MgCl_2$, 0.2 μM of each primer, 1 unit of *Taq* DNA polymerase, and 2.5μL buffer (0.2 M Tris PH. 9.5; 0.25 M KCl; 1 mg/mL BSA, 5 μL/mL Tween 20) in a total volume of 25μL. PCR amplification was performed in a Mastercycler gradient thermal cycler (Eppendorf, Westbury, New York, USA) at 94°C for 60 s, 55°C for 30 s, and 72°C for 60 s for 35 cycles. The residual primers and dNTPs were dephosphorylated using 0.5 unit of

Shrimp Alkaline Phosphatase (SAP, Fermentas, Burlington, Ontario, Canada) and 20 units of Exonuclease I from *E. coli* (Fermentas, Burlington, Ontario, Canada) at 37°C for 2 h. Enzymes were inactivated at 80°C for 20 min.

3. 2. 4 Single nucleotide primer extension reaction and genotyping.

For each candidate SNP, an unlabeled oligonucleotide primer with its 3′ end adjacent to the SNP (Table 3. 3) was synthesized (Operon Biotechnologies, Huntsville, Alabama, USA). A single nucleotide primer extension reaction was performed using an ABI SnaPshot™ Multiplex Ready Reaction Kit (Applied Biosystems, Foster City, California, USA) and 0.2 μM of genotyping primer. The thermal cycling was carried out on an Eppendorf Mastercycler gradient thermal cycler at 95°C for 10 s, 50°C for 5 s, and 60°C for 30 s for 40 cycles. Unincorporated ddNTP was removed from the SnaPshot™ PCR product by adding 1 unit of Shrimp Alkaline Phosphatase (SAP) and incubating at 37°C for 1 h followed by 20 min at 80°C to inactivate the enzyme. Purified extension products were genotyped using an ABI 310 automated genetic analyzer (Applied Biosystems, Foster City, California, USA) and data were processed and analysed using GeneScan software (Applied Biosystems, Foster City, California, USA). Expected SNP genotypes for *P. deltoides* and *P. balsamifera* and sequences of SNP genotyping primers are listed in Table 3. 3.

3. 3 RESULTS

3. 3. 1 Validation of SNPs

The genotypes of all *P. deltoides* and *P. balsamifera* individuals were consistent with the expected genotypes for all nuclear and chloroplast SNPs tested in this study, as listed in Table 3. 3, confirming the reliability and sensitivity of the SNPs for the present study. The nuclear and chloroplast SNP genotypes of the parental individuals used in the controlled crossing experiment were also consistent with the genotypes of the *P. deltoides* and *P. balsamifera* individuals tested in this study. The nuclear SNP genotypes of all *P.* x *jackii* from controlled crosses showed SNP alleles attributable to *P. deltoides* and *P. balsamifera* proving the Mendelian inheritance pattern of these SNPs. The chloroplast genotypes of all control-crossed *P.* x *jackii* were consistent with the chloroplast genotype of *P. deltoides* and this confirmed the maternal inheritance of the chloroplast genome in *Populus*.

3. 3. 2 Genotype of natural P. x jackii

The nuclear and chloroplast genotypes of all 23 natural *P.* x *jackii* individuals tested were consistent with the genotypes of control-crossed *P.* x *jackii*, confirming that these were hybrids between *P. deltoides* and *P. balsamifera*. The morphology (broadly ovate leaves with heart shaped bases) and molecular identification of natural hybrids were congruent, suggesting that the natural hybrid *P.* x *jackii* could be reliably identified using morphological features. All putative natural hybrid individuals tested had nuclear alleles representing both *P. deltoides* and *P. balsamifera* as parental taxa, and the

chloroplast genotype corresponded to that of *P. deltoides*, suggesting an asymmetrical pattern of hybridization in the natural hybrids. Thus, our results clearly show that *P. deltoides* and *P. balsamifera* serve as ovule and pollen donors, respectively, in the *P. x jackii* hybrid complex.

3. 4 DISCUSSION

All species of the genus *Populus* are dioecious and therefore cross-pollination through wind-dispersed pollen is needed for successful fruit set in *P. balsamifera* and *P. deltoides* (Dickmann and Stuart 1983). These two species are somewhat reproductively isolated due to different flowering times. Flowering of *P. balsamifera* generally occurs in April to May and *P. deltoides* flowers from late February to April in the southern distribution range of the species in the United States (Schreiner 1974). In the Oldman river region of southern Alberta, both of these species and their hybrids are known to flower during the month of May (Gom and Rood 1999). Flowering and pollen release may vary by as much as a month among trees in a stand (Farmer 1966; Gom and Rood 1999) and seasonal climatic fluctuations may shift the timing of flowering, leading to phenological overlap between these two species. Thus, some individuals of these two species can hybridize under natural conditions (Brayshaw 1965; Eckenwalder 1984b; Floate 2004).

The observed pattern of natural hybridization between *P. deltoides* and *P. balsamifera*, suggests an asymmetry in the reproductive isolation of these two species. The asymmetric reproductive barriers in general could be attributable to pre- and post-zygotic barriers. Various pre-zygotic mechanisms that may cause asymmetry in hybridization success include differences in style length (Kiang and Hamrick 1978; Sorenson and Brewbaker 1994), differential fruit abortion (Levin 1978; Howard et al. 1998), and self incompatibility systems (Lewis and Crowe 1958). Controlled crosses involving female *P. deltoides* and male *P. balsamifera* showed a high success rate and

produced viable seeds, whereas reciprocal crosses involving female *P. balsamifera* and male *P. deltoides* produced only a few seeds and the viability of these seeds was very low. For instance, in a crossing experiment performed in 1998, no seed set was observed in all 20 crossings between female *P. balsamifera* and male *P. deltoides*, whereas five out of 16 crosses between female *P. deltoides* and male *P. balsamifera* produced viable progenies. In another crossing experiment performed in 2004, only two out of 19 crosses between female *P. balsamifera* and male *P. deltoides* produced seeds, giving rise to only one seedling. Conversely, four out of 30 crosses between female *P. deltoides* and male *P. balsamifera* produced seeds and gave rise to over 600 seedlings. Similarly, the controlled crosses between female *P. trichocarpa*, and male *P. deltoides* yield seeds with low viability, commonly associated with premature dehiscence of the capsule. Seedlings of these crosses are routinely produced through embryo rescue techniques (Dickmann 2001; Riemenschneider et al. 2001) suggesting a post-zygotic incompatibility between these two species. *P. trichocarpa* is evolutionarily closely related to *P. balsamifera* (Hamzeh and Dayanandan 2004), and therefore, it is logical to assume that the asymmetric reproductive isolation observed in the *P. deltoides* and *P. balsamifera* complex may be mediated through post-zygotic mechanisms. The evidence for asymmetric cross incompatibility has also been found in a *P. angustifolia* and *P. fremontii* hybrid complex in Utah. Restriction fragment length polymorphisms (RFLP) analysis in this hybrid complex showed that the hybrid population consisted of either F_1 hybrids or backcrosses to *P. angustifolia* (Keim et al. 1989). No trees attributable to crosses between F_1 or between F_1 and *P. fremontii* were found. The controlled backcrossing of F_1 hybrids to *P.*

fremontii resulted in early death of seedlings, suggesting a post-zygotic developmental incongruity (Keim et al. 1989; Hogenboom 1984).

We speculate that the observed asymmetries in hybridization between *P. deltoides* and *P. balsamifera* may be due to cyto-nuclear incompatibility resulting from divergent evolution of nuclear and cytoplasmic genes coding for proteins interacting in photosynthesis or respiration that affect the viability of hybrids (Michaelis 1954; Levin 1978; Wu et al. 1999). Alternatively, the cytonuclear incompatibility may result from the activity of transposable elements, which are abundant in many plant genomes (Tiffin et al. 2001). The activity of some transposable elements can be suppressed by maternally inherited factors (Engels 1989). Therefore, hybrid progeny with a cytoplasmic background that has not evolved with a transposable element may experience greater transposon activity leading to cytonuclear genetic incompatibility and reduced hybrid fitness or viability. Further studies focused on cyto-nuclear interactions in the *P . x jackii* hybrid complex would be valuable to gain insights into mechanisms associated with asymmetric gene flow in forest trees.

In theoretical models of reproductive barrier reinforcement, the reciprocal crosses between lineages are assumed to be equally compatible (Felsenstein 1980; Liou and Price 1994; Kelly and Noor 1996). However, the asymmetries in gene flow between partially isolated taxa may greatly reduce the probability of evolution of reinforcing reproductive barriers (Servedio and Kirkpatrick 1997) and lineages that exhibit asymmetries in reproductive isolation are more likely to introgress than evolve reinforcement. Under these circumstances introgression is likely to be directional, with genes moving mainly from one species to another, and to affect the phenotypic and genotypic variation in

natural populations (Stebbins 1959). The proportion of parental genomes in hybrid species may serve as an indicator for assessing the directionality of introgression (Martinsen et al. 2001) and cytoplasmic and nuclear interactions in the hybrid zone (Rieseberg 1995).

In summary, our findings confirm the presence of asymmetric reproductive isolation in two inter-fertile plant species under natural conditions. Our data, based on both the biparentally-inherited nuclear and the maternally-inherited chloroplast genome, provide direct evidence for the long suspected presence of asymmetries in interspecific crossing barriers in *Populus* under natural conditions.

Table 3. 1 Individuals of *P. deltoides*, *P. balsamifera*, and *P. x jackii* natural stands sampled for this study, voucher numbers, source and reference numbers, and geographic origin where available; AP, Alberta Pacific Forest Products Collection; MRNFQ, Le Ministere des Ressources Naturelles et de la Faune du Quebec.

Taxon	Voucher number	Source and Reference number	Sample's origin
P. deltoides Bartr. ex Marsh.	MH-048	AP 145 / 3277	Bindloss/Alberta
	MH-049	AP 153 / 3285	Bindloss/Alberta
	MH-050	AP 154 / 3286	Bindloss/Alberta
	MH-053	MRNFQ / Native	Leclercville-Rive du Chene/Quebec
	MH-119	AP 181 / 3355	Policepoint
	MH-120	AP 25	
	MH-121	AP 140	Bowisland
	MH-122	AP 159 / 3291	
	MH-123	AP 170 / 3302	Dinosaurpark
	MH-124	AP 178 / 3311	Policepoint
	MH-126	MRNFQ / 4118	Lotbiniere/Quebec
	MH-127	MRNFQ / 4380	Lotbiniere/Quebec
	MH-128	MRNFQ / 4403	Lotbiniere/Quebec
	MH-129	MRNFQ / 4413	Lotbiniere/Quebec
	MH-130	MRNFQ	Leclercville-Rive du Chene/Quebec
	MH-131	MRNFQ	
	MH-132	MRNFQ	
	MH-133	MRNFQ	
	MH-134	MRNFQ	
	MH-135	MRNFQ	
	MH-136	MRNFQ	
	MH-137	MRNFQ	
	MH-138	MRNFQ	
	MH-139	MRNFQ	
	MH-140	MRNFQ	
	MH-141	MRNFQ	
	MH-142	MRNFQ	
	MH-143	MRNFQ	
	MH-144	MRNFQ	
	MH-145	MRNFQ	
P. balsamifera L.	MH-054	AP 64 / pb# 12-93	Smith / Alberta
	MH-055	AP 65 / pb# 13-93	Salteaux River / Alberat
	MH-056	AP 66 / pb# 14-93	Ctaonklin Winter Road/ Alberta
	MH-080	AP 68 / pb# 16	Poplarcreel
	MH-081	AP 70 / pb# 18	Fort Mcmur
	MH-083	AP 56 / pb# 4	Wabasca
	MH-109	Natural stands	Edmonton/Alberta
	MH-110	Natural stands	Edmonton/Alberta
	MH-111	Natural stands	Edmonton/Alberta
	MH-112	Natural stands	Edmonton/Alberta

P .x jackii Sarg.	NJ-1	MRNFQ / 16	Ville de Quebec
	NJ-2	MRNFQ / 22	Charlebourg
	NJ-3	MRNFQ / 70	Ville de Quebec
	NJ-4	MRNFQ / 315	St Bruno de Montarville
	NJ-5	MRNFQ / 316	St Bruno de Montarville
	NJ-6	MRNFQ / 410	Ste Francoise de Lotbiniere
	NJ-7	MRNFQ / 1005	Ville de Quebec
	NJ-8	MRNFQ / 1012	Ville de Quebec
	NJ-9	MRNFQ / 1061	Portneuf
	NJ-10	MRNFQ / 1063	Portneuf
	NJ-11	MRNFQ / 1064	Portneuf
	NJ-13	MRNFQ / 1078	Portneuf
	NJ-14	MRNFQ / 1081	Portneuf
	NJ-15	MRNFQ / 1082	Portneuf
	NJ-16	MRNFQ / 1083	Portneuf
	NJ-19	MRNFQ / 1170	St Redempteur Levis
	NJ-20	MRNFQ / 3057	Ontario
	NJ-36	AP 8 // MRNFQ-1014	Quebec City
	NJ-37	AP 9 // MRNFQ-1016	Quebec City
	NJ-38	AP 10 // MRNFQ- 1017	Quebec City
	NJ-39	MRNFQ / 1062	Portneuf
	NJ-40	MRNFQ / 1079	Portneuf
	NJ-41	MRNFQ / 1080	Portneuf
	NJ-42	MRNFQ / 1081	Portneuf

Table 3. 2 Parental Clones of controlled crosses of *P.x jackii* and individuals of controlled-crossed *P. x jackii* sampled for this study, voucher numbers, source and reference numbers of the parental clones and hybrid family number of controlled crosses indicating their parental clones numbers. Leaf or bud material for pollen parent clones 8-2004 and 12-2004 were not available.

Taxon	Voucher number	Source and Reference number
Parental Clones of controlled crosses of *P.x jackii*		
Seed parent, *P. deltoides*	MCJ 1	4954 / f 10224-2
	MCJ 2	4942 / f 10220-9
Pollen parent, *P. balsamifera*	FCJ-1	a-172
Controlled crosses of *P. x jackii*		Hybrid Family number (seed P. # x Pollen p.#)
	CJ-1	23467 [4942 (MCJ-2) X 12-2004]
	CJ-2	23467 [4942 (MCJ-2) X 12-2004]
	CJ-3	23467 [4942 (MCJ-2) X 12-2004]
	CJ-4	23467 [4942 (MCJ-2) X 12-2004]
	CJ-5	23467 [4942 (MCJ-2) X 12-2004]
	CJ-12	23158 [4954 (MCJ-1) X 8-2004]
	CJ-13	23158 [4954 (MCJ-1) X 8-2004]
	CJ-14	23158 [4954 (MCJ-1) X 8-2004]
	CJ-15	23158 [4954 (MCJ-1) X 8-2004]
	CJ-16	23158 [4954 (MCJ-1) X 8-2004]
	CJ-26	23509 [4942 (MCJ-2) X a-172 (FCJ-1)]
	CJ-27	23509 [4942 (MCJ-2) X a-172 (FCJ-1)]
	CJ-28	23509 [4942 (MCJ-2) X a-172 (FCJ-1)]
	CJ-29	23509 [4942 (MCJ-2) X a-172 (FCJ-1)]
	CJ-30	23509 [4942 (MCJ-2) X a-172 (FCJ-1)]
	CJ-31	23509 [4942 (MCJ-2) X a-172 (FCJ-1)]
	CJ-32	23509 [4942 (MCJ-2) X a-172 (FCJ-1)]
	CJ-33	23509 [4942 (MCJ-2) X a-172 (FCJ-1)]
	CJ-34	23509 [4942 (MCJ-2) X a-172 (FCJ-1)]
	CJ-35	23509 [4942 (MCJ-2) X a-172 (FCJ-1)]

Table 3. 3 The single nucleotide polymorphism (SNP) locus, genotypes in *P. deltoides* *P. balsamifera* and *P.* x *jackii*, and genotyping primer sequences of nuclear and chloroplast SNPs used in the present study. The nrSNP1 and nrSNP2 are located within the ITS2 region of the nuclear rDNA. The cpSNP1 is located within the intergenic region of *trnT-trnL*, and cpSNP2, cpSNP3, and cpSNP4 are located in the *trnL* intron of the chloroplast genome.

SNP locus	Genotype			Primer name and sequence
	P. deltoides	*P. balsamifera*	*P. x jackii*	
nrSNAP 1	GG	AA	GA	SNPBLIT2R: 5'-CGTGAGCCGAGGGGAG-3'
nrSNAp 2	TT	CC	TC	SNPBLIT3R: 5'-GATGCCCGAGGGTCC-3'
cpSNP 1	A	T	A	PBLIG1F: 5'-ACGTGAAATGAATATAGAAAAATGAATTGAATATA-3'
cpSNP 2	G	T	G	SNPDLTN1F: 5'-TAAAGGAATCCTTCTGTTAAAGT-3'
cpSNP 3	C	T	C	SNPBLTN1F: 5'-ACCCTATAAACATAATACATAGGAAA-3'
cpSNP 4	A	T	A	PBLTN2R: 5'-ACGTACGTACGTACGTACGTACGATTCAAATCAAAGCAATTTT-3'

GENERAL CONCLUSION

Nuclear genome based ISSR markers and six combined nuclear genome based SCISSR data sets were used to assess genetic relationships among species of the genus *Populus*, a closely related group of species, in which other genetic markers showed limited success in resolving the evolutionary relatedness. The phylogenetic tree based on seven ISSR markers was congruent with existing phylogenetic trees of poplars based on other molecular and morphological data, but showed higher resolution than RFLP and DNA sequence based phylogenetic trees. The phylogenetic tree based on six combined SCISSR markers showed higher resolution than RFLP, ISSR, single SCISSR, and DNA sequence based phylogenetic trees. However, most of the internal nodes depicting speciation events (and therefore, phylogenetic relationships among species) either were not resolved or supported by bootstrap analysis or test of credibility.

Based on this analysis species of section *Populus* as well as North American cottonwoods of section *Aigeiros* (or in other words, species of section *Aigeiros* with the exception of *P. nigra*) proved to be of monophyletic origin, whereas species of section *Tacamahaca* appeared to have a paraphyletic origin. However, this was only due to the placement of *P. nigra* in the clade including three Asiatic members of section *Tacamahaca*. Otherwise, section *Tacamahaca* would also have a monophyletic origin. For the first time ISSR and SCISSR based phyligenetic analysis revealed the nuclear genomic affinity of *P. nigra* to the members of section *Tacamahaca* and more precisely to the Asiatic members of the section. The close genetic relationship between *P. nigra* and species of section *Tacamahaca* also agrees with evidence from various phenotypic

traits, interfertility and the chemistry of bud exudates and serves as evidence for introgression between *P. nigra* and species of section *Tacamahaca*.

The results of this study as well as a previous one (Hamzeh and Dayanandan 2004) are good evidence that analyses based on a single or a small number of genes or genomic data provide insufficient evidence for establishing phylogenetic hypotheses. It is only through analyses of a large amount of sequence data that a robust phylogenetic reconstruction can be obtained. Therefore, in order to gain insight into the evolutionary history of species of the genus *Populus* we need larger concatenated genome-wide sampling of independently evolving sequences for the precise reconstruction of the phylogenetic association of all the species.

Furthermore, both nuclear and chloroplast based SNPs specific for *P. deltoides* and *P. balsamifera* were used to test the hybrid status of morphologically based putative hybrids, and determine the maternal and paternal parental species of *P.* x *jackii* (F_1 hybrids between these two species) individuals collected from nature to infer the direction of hybridization in natural populations. The result of this study confirmed the presence of asymmetrical reproductive isolation between two inter-fertile plant species under natural conditions. Data based on both bi- parentally inherited nuclear and maternally inherited chloroplast genome provided direct evidence for the long suspected presence of asymmetries in interspecific crossing barriers in *Populus* under natural conditions.

BIBLIOGRAPHY

ANDERSON, E. 1949. Introgressive hybridization. John Wiley and Sons, New York, New York, USA.

ANDERSON, E. 1948. Hybridization of the habitat. *Evolution* 2: 1-9.

ANDERSON, E., AND G. L., STEBBINS. 1954. Hybridization as an evolutionary stimulus. *Evolution* 8: 378-388.

ANDERSON, E., AND L. HUBRICHT. 1938. Hybridization in *Tradescantia*. III. The evidence for introgressive hybridization. *American Journal of Botany* 25: 396-402.

ARNOLD. M. L. 1997. Natural hybridization and evolution. Oxford University Press. New York, USA.

ARNOLD, M. L.1993. *Iris nelsonii* (Iridaceae): origin and genetic composition of a homoploid hybrid species . *American Journal of Botany* 80: 577-583.

ARNOLD, M. L.1992. Natural hybridization as an evolutionary process. *Annual Review of Ecology and Systematics* 23: 237-261.

AZUMA, T., T. KAJITA, J. YOKOYAMA, AND H. OHASHI. 2000. Phylogenetic relationships of *Salix* based on *rbcL* sequence data. *American Journal of Bot*ony 87: 67-75.

BALDAUF, S. L., A. J. ROGER, I. WENK-SIEFERT, AND W. F. DOOLITTLE. 2000. A kingdom-level phylogeny of eukaryotes based on combined protein data. *Science* 290: 972–977.

BALDAUF, S. L. AND J. D. PALMER. 1993. Animals and fungi are each other's closest relatives: congruent evidence from multiple proteins. *Proceedings of the National Academy of Sciences, USA* 90: 11558–11562.

BARNES, B. V. 1961. Hybrid aspens in the lower peninsula of Michigan. *Rhodora* 63: 311-324.

BARNES, B. V., AND K. S. PREGITZER. 1985. Occurrence of hybrids between bigtooth and trembling aspen in Michigan. *Canadian Journal of Botany* 63: 1888-1890.

BARRETT, J., O.P. RAJORA, F. C. H. YEH, AND B. P. DANCIK. 1993. Mitochondrial DNA variation and genetic relationships of *Populus* species. *Genome* 36: 87-93.

BECCERRA, J., AND VENABLE, D. L. 1999. Nuclear ribosomal DNA phylogeny and its implications for evolutionary trends in Mexican *Bursera*. *American Journal of Botany* 86: 1047–1057.

BERGTHORSSON, U., K. L. ADAMS, B. THOMASON, AND J. D. PALMER. 2003. Widespread horizontal transfer of mitochondrial genes in flowering plants. *Nature* 424: 197–201.

BORNET, B. AND M. BRANCHARD. 2001. Non-anchored inter simple sequence repeats (ISSR) markers: reproducible and polymorphic tools for genome fingerprinting. *Plant Molecular Biol*ogy 19: 209-215.

BRAATNE, J. H., T. M. HINCKLEY, AND R. F. STETTLER. 1992. Influence of soil water supply on the physiological and morphological components of plant water balance in *Populus trichocarpa, Populus deltoides* and their F1 hybrids. pp. 57-85. *In* R. F. Stettler, H. D. Bradshaw, Jr., P. E. Heilman, and T. M. Hinckley [eds.], Biology of *Populus* and its Implications for Management and Conservation. Part I, Chapter 3. NRC Research Press, National Research Council of Canada, Ottawa, ON, Canada.

BRADSHAW, H. D. JR., R. CEULEMANS, J. DAVIS, AND R. STETTLER. 2000. Emerging Model Systems in Plant Biology: Poplar (*Populus*) as a Model Forest Tree. *Journal of Plant Growth Regulation* 19: 306-313.

BRAYSHAW, T. C. 1965. Native poplars of southern Alberta and their hybrids. Canadian Department of Forestry, Ottawa. Publ. 1109.

BULL, J. J., J. P. HUELSENBECK, C. W. CUNNINGHAM, D. L. SWOFFORD, AND P. J. WADDELL. 1993. Partitioning and Combining Data in Phylogenetic Analysis. *Systematic Biology* 42: 384-397.

BURKE, J. M., AND M. L. ARNOLD. 2001. Genetics and Fitness of hybrids. *Annual Review of Genetics* 35: 31-52

CARROLL, S. B., J. K. GRENIER, AND S. D. WEATHERBEE. 2001. From DNA to diversity. Blackwell Science, Oxford, UK.

CHO, R. J., MINDRINOS, M., RICHARDS, D. R., SAPOLSKY, R. J., ANDERSON, M., DRENKARD, E., DEWDNEY, J., REUBER, T. L., STAMMERS, M., FEDERSPIEL, N., THEOLOGIS, A., YANG, W.H., HUBBELL, E., AU, M., CHUNG, E. Y., LASHKARI, D., LEMIEUX, B., DEAN, C., LIPSHUTZ, R. J., AUSUBEL, F. M., DAVIS, R. W., AND OEFNER, P. J. 1999. Genome-wide mapping with biallelic markers in *Arabidopsis thaliana*. *Nature Genetics* 23: 203-207.

CULLING, K. W. 1992. Design and testing of plant–specific PCR primer for ecological and evolutionary studies. *Molecular Ecology* 1: 223–240.

CUMMINGS, M. P., S. P. OTTO, AND J. WAKELEY. 1995. Sampling properties of DNA sequence data in phylogenetic analysis. *Molecular and Biological Evolution* 12: 814–822.

CUNNINGHAM, C. W.1997. Can three incongruence tests predict when data should be combined? *Molecular Biology and Evolution* 14: 733-740.

DAYANANDAN, S., K. S. BAWA, AND R. V. KESSELI. 1997. Conservation of microsatellite among tropical trees (*leguminosae*). *American Journal of Botany* 84: 1658-1663.

DE QUEIROZ, A. 1993. For consensus(sometimes). *Systematic biology* 42: 368-372.

DE QUEIROZ, A., M. J. DONOGHUE, AND J. KIM. 1996. Separate versus combined analysis of phylogenetic evidence. *Annual Review of Ecology and Systematics* 26: 657-681.

DICKMANN, D. I. 2001. An overview of the genus *Populus*. *In* Poplar Culture in North America. *Edited by* D.I. Dickmann, J.G. Isebrands, J.E. Eckenwalder, and J. Richardson. NRC Research Press, Ottawa, Ontario. pp. 1-42.

DICKMANN, D. I., J. G. ISEBRANDS, J. E. ECKENWALDER, AND J. RICHARDSON. 2001. Poplar culture in North America. NRC Research Press, Ottawa, Ontario, Canada.

DICKMANN, D. I., AND K. STUART. 1983. The culture of poplars in Eastern North America. Department of Forestry, Michigan State University, East Lansing, Michigan, USA.

DOBZHANSKY, T. H. 1940. Speciation as a stage in evolutionary divergence. *American Naturalist* 74: 312 321.

DOYLE, J. J. AND J. L. DOYLE. 1987. A rapid DNA isolation procedure for small quantities of fresh
leaf tissue. *Phytochemical Bulletin*. 19: 11-15.

ECKENWALDER, J. E. 1996. Systematics and evolution of *Populus*. *In* R. F. Stettler, H. D.

Bradshaw, Jr., P. E. Heilman, and T. M. Hinckley [eds.], Biology of *Populus* and its implications for management and conservation, 7–32. NRC Research Press, National Research Council of Canada, Ottawa, Ontario, Canada.

ECKENWALDER, J. E. 1984a. Natural intersectional hybridization between North American species of *Populus* in sections *Aigeiros* and *Tacamahaca*. I. Population studies of *P. parryi*. *Canadian Journal of Botany* 62: 317-324.

ECKENWALDER, J. E. 1984b. Natural intersectional hybridization between North American species of *Populus* in sections *Aigeiros* and *Tacamahaca*. II. Taxonomy. *Canadian Journal of Botany* 62: 325-335.

ECKENWALDER, J. E. 1984c. Natural intersectional hybridization between North American species of *Populus* in sections *Aigeiros* and *Tacamahaca*. III. Paleobotany and evolution. *Canadian Journal of Botany* 62: 336-342.

ECKENWALDER, J. E. 1982. *Populus* x *inopia* hybr. nov. (Salicaceae), a natural hybrid between the native North American *P. fremontii* S. Watts and the introduced Eurasian *P. nigra* L. *Madrono* 29: 67-78.

ECKENWALDER, J. E. 1977a. North American cottonwoods (*Populus*, Salicaceae) of sections *Abaso*, and *Aigeiros*. *Journal of the Arnold Arboretum* 58: 193-208.

ECKENWALDER, J. E. 1977b. Systematics of *Populus L.* in southwestern North America with special reference to section *Aigeiros* Duby. Ph.D thesis, University of California, Berkeley, California, USA

EERNISSE, D. J. AND A. G. KLUGE. 1993. Taxonomic congruence versus total evidence, and amniote phylogeny inferred from fossils, molecules, and morphology. *Molecular Biology and Evolution* 10: 1170-1195.

ELLSTRAND, N. C., AND ELAM, D. R. 1993. Population genetic consequences of small population size: implications for plant conservation. *Annual Review of Ecology and Systematics* 24: 217–242.

ENGELS, W. R. 1989. P elements in *Drosophila*. *In* Mobile DNA. *Edited by* D. Berg and M. Howe. American Society of Microbiology Press, Washington, DC. pp. 437-484.

FARMER, R. E., JR. 1966. Variation in time of flowering and seed dispersal of eastern cottonwood in the lower Mississippi Valley. *Forest Science* 12: 343-347.

FARRIS, J. S., M. KALLERSJO, A. G. KLUGE, AND C. BULT. 1995. Constructing a Significance Test for Incongruence. *Systematic Biology* 44: 570-572.

FARRIS, J. S., M. KALLERSJO, A. G. KLUGE, AND C. BULT. 1994. Testing significance of incongruence. *Cladistics* 5: 417-419.

FELSENSTEIN, J. 1985. Phylogenies and comparative method. *American Naturalist* 125: 1-

15.

FELSENSTEIN, J. 1980. Skepticism towards Santa Rosalia or why are there so few kinds of animals? *Evolution* 35: 124-138.

FLOATE, K. D. 2004. Extent and patterns of hybridization among the three species of *Populus* that constitute the riparian forest of southern Alberta, Canada. *Canadian Journal of Botany* 82: 253-264.

FOWELLS, H. A. 1965. Silvics of forest trees of the United States. US Department of Agriculture, Forest Service. Agriculture Handbook. Washington D. C., USA.

FUTUYMA, D. J. 1998. Evolutionary biology. Sinauer Associates, Sunderland,

Massachusetts, USA.

GIRIBET, G., G. D. EDGECOMBE, AND W. C. WHEELER. 2001. Arthropod phylogeny based on eight molecular loci and morphology. *Nature* 413: 157–161.

GOM, L. A., AND ROOD, S. B. 1999. The discrimination of cottonwood clones in a mature grove along the Oldman River in southern Alberta. *Canadian Journal of Botany* 77: 1084-1094.

GRANT, V. 1981. Plant speciation. Columbia University Press, New York, New York.

GRANT, V. 1953. The role of hybridization in the evolution of leafy-stemmed gilias. *Evolution* 7: 51-64.

GRANT, P. R., AND GRANT, B. R. 1996. High survival of Darwin's finch hybrids: Effects of beak morphology and diets. *Ecology* 77: 500-509.

GRANT, P. R., AND GRANT, B. R. 1994. Phenotypic and genetic effects of hybridization in Darwin's finches. *Evolution* 48: 297-316.

GRAYBEAL, A. 1998. Is it better to add taxa or characters to a difficult phylogenetic problem? *Systematic Biology* 47: 9–17.

GREENAWAY, W., ENGLISH, S., WHATLEY, F. R., AND ROOD, S. B. 1991. Interrelationships of poplars in a hybrid swarm as studied by gas chromatography-mass spectrometry. *Canadian Journal of Botany* 69: 203-208.

GRIFFIN, T. J., AND SMITH, L. M. 2000. Single-Nucleotide Polymorphism Analysis by MALDI-TOF Mass Spectrometry. Trends Biotechnol18: 77-84.

GUPTA, M., Y. S. CHYI, J. ROMERO-SEVERSON, AND J. L. OWEN. 1994. Amplification of DNA markers from evolutionary diverse genomes using single primers of simple sequence repeats. *Theoretical and Applied Genetics* 89: 998-1006.

HAMZEH, M., P. PÉRINET AND S. DAYANANDAN. 2006. Genetic relationships among species of *Populus* (Salicaceae) based on nuclear genomic data. *Journal of the Torrey Botanical Society* 133: 519–527.

HAMZEH, M., AND S. DAYANANDAN. 2004. Phylogeny of *Populus* (Salicaceae) based on nucleotide sequences of chloroplast *trnT–trnF* region and nuclear rDNA. *American Journal of Botany* 91: 1398–1408.

HARRISON, R. G. 1990. Hybrid zones: windows on evolutionary process. *Oxford Surveys in Evolutionary Biology*, Oxford University Press.

HART, M. W., AND J. SUNDAY. 2007. Things fall apart: biological species form unconnected parsimony networks. *Biology Letters* 3: 509-512.

HEILMAN, P. E. AND R. F. STETTLER. 1985. Genetic variation and productivity of *Populus trichocarpa* T. & G. and its hybrids. II. Biomass productivity in a four year plantation. *Canadian Journal of Foret Resources* 15: 384-388

HEISER, C. B. JR. 1949. Natural hybridization with particular reference to introgression. *Botanical Review* 15: 645-687.

HOGENBOOM, N. G. 1984. Incongruity: Non-functioning of intercellular and intracellular partner relationships through non-matching information. *In* Cellular interactions. *Edited by* H.F. Linskens, and J. Heslope-Harrison. Springer-Verlag, Berlin. pp. 640-654.

HOWARD, D. J. 1993. Reinforcement: origin, dynamics and fate of an evolutionary hypothesis. *In* Hybrid zones and the evolutionary process. *Edited by* R. G. Harrison. Oxford University Press, New York, New York. pp. 46-69.

HOWARD, D. J., REECE, M., GREGORY, P. G., CHU, J., AND CAIN, M. L. 1998. The evolution of barriers to fertilization between closely related organisms. *In* Endless forms: species and speciation. *Edited by* D. J. Howard and S. H. Berlocher. Oxford University Press, New York, New York. pp. 279-290.

HUELSENBECK, J. P. 1995. Performance of phylogenetic methods in simulation. *Systematic Biology* 44: 17–48.

HUELSENBECK J. P., AND F. RONQUIST. 2001. MrBayes: Bayesian inference of phylogeny. *Bioinformatics* 17: 754-755.

HUELSENBECK, J. P., F. RONQUIST, R. NIELSEN, AND J. P.BOLLBACK. 2001. Evolution – Bayesian inference of phylogeny and its impact on evolutionary biology. *Science* 294: 2310–2314

HUELSENBECK J. P., J. J. BULL, AND C. W. CUNNINGHAM. 1996. Combining data in phylogenetic analysis. *Trends in Ecology and Evolution* 11: 152-158.

HWANG, U. W., M. FRIEDRICH, D. TAUTZ, C. J. PARK, AND W. KIM. 2001. Mitochondrial protein phylogeny joins myriapods with chelicerates. *Nature* 413: 154–157.

KEIM, P., PAIGE, K. N., WHITHAM, T.G., AND LARK, K.G. 1989. Genetic analysis of an interspecific hybrid swarm of *Populus*: occurrence of unidirectional introgression. *Genetics* 123: 557-565.

KELLY, J. K., AND NOOR, M. A. 1996. Speciation by reinforcement: a model derived from studies of *Drosophila*. *Genetics* 143: 1485-1497.

KIANG, Y. T., AND HAMRICK, J. L. 1978. Reproductive isolation in the *Mimulus guttatus M. nasutus* complex. *American Midland Naturalist* 100: 269-276.

KLUGE, A. G. 1998. Total Evidence Or Taxonomic Congruence: Cladistics Or Consensus

Classification. *Cladistics* 14: 151–158.

KLUGE, A. G. 1989. A Concern for Evidence and a Phylogenetic Hypothesis of Relationships Among Epicrates (Boidae, Serpentes). *Systematic Zoology* 38: 7-25.

KNOBLOCH, I. W. 1971. Intergeneric hybridization in flowering plants. *Taxon* 21: 97–103.

KOJIMA, T., T. NAGAOKA, AND Y. OGIHARA. 1998. Genetic linkage map of ISSR and RAPD markers in Einkorn wheat in relation to that of the RFLP markers. *Theoretical and Applied Genetics* 96: 37-45.

KOPP, A. AND J. R. TRUE. 2002. Phylogeny of the Oriental Drosophila melanogaster species group: a multilocus reconstruction. *Systematic Biology* 51: 786–805.

LESKINEN, E. AND C. ALSTROME-RAPAPORT. 1999. Molecular phylogeny of Salicaceae and closely related Flacourtiaceae: evidence from *5.8S*, *ITS1* and *ITS2* of the rDNA. *Plant Systemetics and Evolution* 215: 209-227.

LEVIN, D. A. 1978. The origin of isolating mechanisms in flowering plants. Evol. Biol. 11: 185-315.

LEVIN, D. A, FRANCISCO-ORTEGA, J., AND JANSEN, R. K. 1996. Hybridization and the extinction of rare species. *Conservation Biology* 10: 10–16.

LEWIS, D., AND CROWE, L. K. 1958. Unilateral interspecific incompatibility in flowering plants. *Heredity* 12: 233-256.

LEWONTIN, R. C., AND BIRCH, L. C. 1966. Hybridization as a source of variation for adaptation to new environments. *Evolution* 20: 315-336.

LIBERLES, D. A., D. R. SCHREIBER, S. GOVINDARAJAN, S. G. CHAMBERLIN, AND S. A. BENNER. 2001. The adaptive evolution database (TAED). *Genome Biology* 2: 1-6.

LIOU, L. W., AND PRICE, T. D. 1994. Speciation by reinforcement of premating isolation. *Evolution* 48: 1451-1459.

LITTLE, E. L. 1979. Checklist of United States trees. U. S. For. Serv. Agric. Handbook. No. 541.

LITTLE, E. L. 1971. Atlas of United States trees, volume 1, conifers and important hardwoods. U.S. Department of Agriculture, Miscellaneous Publication 1146. Washington, D.C. USA.

LIU, F-G. R. AND M. M. MIYAMOTO. 1999. Phylogenetic Assessment of Molecular and Morphological Data for Eutherian Mammals. *Systematic Biology* 48:54-64.

LOYTYNOJA, A. AND M. C. MILINKOVITCH. 2001. Molecular phylogenetic analyses of the mitochondrial ADP–ATP carriers: the Plantae/Fungi/Metazoa trichotomy revisited. *Proceedings of the National Academy of Sciences, USA* 98: 10202–10207.

MADDISON, W. P. 1997. Gene trees in species trees. *Systematic Biology* 46: 523–536.

MADDISON, W. P. 1990. A method for testing the correlated evolution of two binary characters: are gains or losses concentrate on certain branches of a phylogenetic tree? *Evolution* 44: 304-314.

MADDISON, D. R., AND W. P. MADDISON. 2001. MacClade 4. Sinauer Associate. Sunderland, Massachusetts, USA.

MARTIN, A. P. AND T. M. BURG. 2002. Perils of paralogy: using HSP70 genes for inferring organismal phylogenies. *Systematic Biology* 51: 570–587.

MARTINS, E. P. 1995. Phylogenies and comparative data, a microevolutionary perspective. *Philosophical transactions: Biological Sciences* 349: 85-91.

MARTINSEN, G. D., WHITHAM, T. G., TUREK, R. J., AND KEIM, P. 2001. Hybrid populations selectively filter gene introgression between species. *Evolution* 55: 1325-1335.

MASON-GAMER, R. J. AND E. A. KELLOGG. 1996. Testing for phylogenetic conflict among molecular data sets in the tribe Triticeae (Gramineae). *Systematic Biology* 45: 522–543.

MAYR, E. 1963. Animal species and evolution. Belknap Press, Cambridge, Mass.

MERRITT, T. J., AND J. M. QUATTRO. 2001. Evidence for a period of directional selection following gene duplication in a neurally expressed locus of triosephosphate isomerase. *Genetics* 159: 689-697.

MICHAELIS, P. 1954. Cytoplasmic inheritance in *Epilobium* and its theoretical significance. *Advances in Genetics* 6: 287-401.

MIYAMOTO, M. M. AND W. M. FITCH. 1995. Testing Species Phylogenies and Phylogenetic Methods with Congruence. *Systematic Biology* 44: 64-76.

MOREIRA, D., H. LE GUYADER, H. AND H. PHILIPPE. 2000. The origin of red algae and the evolution of chloroplasts. *Nature* 405: 69–72.

MURPHY, W. J., E. EIZIRIK, W. E. JOHNSON, Y. P. ZHANG, O. A. RYDERK, AND S. J. O'BRIEN. 2001. Molecular phylogenetics and the origins of placental mammals. *Nature* 409: 614–618.

NAUMOV, G. I., S. A. JAMES, E. S. AUMOVA, E. J. LOUIS, AND I. N. ROBERTS. 2000. Three new species in the Saccharomyces sensu stricto complex: Saccharomyces cariocanus, Saccharomyces kudriavzevii and Saccharomyces mikatae.

International Journal of Systematic and Evolutionary Microbiology 50: 1931–1942.

NYLANDER, J. A. A., F. RONQUIST, J. P. HUELSENBECK, AND J. L. NIEVES-ALDREY. 2004. Bayesian phylogenetic inference of combined data. *Systematic Bioogyl* 53: 47–67

OTTO, S. P., AND J. WHITTON. 2000. Polyploid incidence and evolution. *Annual Review of Genetics* 24: 401-437.

PAGE, R. D. M., AND E. C. HOLMES. 1998. Molecular Evolution: a phylogenetic approach. Blackwell Publishing. UK.

POSADA, D. ANDK. A. CRANDALL. 1998. Modeltest: testing the model of DNA substitution. *Bioinformatics* 14: 817–818

PHILIPPE, H., A. CHENUIL, AND A. ADOUTTE. 1994. Can the cambrian explosion be inferred through molecular phylogeny? *Development* 15–25.

RAHMAN, M. H., AND O.P. RAJORA 2002. Microsatellite DNA fingerprinting, differentiation, and genetic relationships of clones, cultivars, and varieties of six poplar species from three sections of the genus *Populus*. *Genome* 45: 1083-1094.

RAJORA, O. P., 1989. Pollen competition among *Populus deltoides* Marsh., *P. nigra* L. and *P. maximowiczii* Henry in fertilizing *P. deltoides* ovules and siring its seed crop. *Sexual Plant Reproduction* 2: 90-96.

RAJORA, O. P., AND B. P. DANCIK. 1995. Chloroplast DNA variation in *Populus*. Interspecific restriction fragment polymorphisms and genetic relationships among *Populus deltoides*, *P. nigra*, *P. maximowiczii*, *P.* x *Canadensis*. *Theoretical and Applied Genetics* 90: 324- 330.

RAJORA, O. P., AND L. ZSUFFA. 1990. Allozyme divergence and evolutionary

relationships among *Populus deltoides*, *P. nigra* and *P. maximowiczii*. *Genome* 33: 44-49.

RANDOLPH L. F. 1966. *Iris nelsonii*, a new species of Louisiana iris of hybrid origin. *Baileya* 14: 143–69.

RIEMENSCHNEIDER, D. E., STANTON, B. J., VALLEE, G., AND PERINET, P. 2001. Poplar breeding strategies. *In* Poplar Culture in North America. *Edited by* D. I. Dickmann, J. G. Isebrands, J. E. Eckenwalder, and J. Richardson. NRC Research Press, Ottawa, Ontario. pp. 43-76.

RIESEBERG, L. H. 1997. Hybrid origins of plant species. *Annual Review in Ecology and Systematics* 28: 359-389.

RIESEBERG, L. H. 1995. The role of hybridization in evolution: old wine in new skins. *American Journal of Botany* 82: 944-953.

RIESEBERG, L. H., J. WHITTON, AND C. R. LINDER. 1996. Molecular marker incongruence in plant hybrid zones and in phylogenetic trees. *Acta Botanica Neerland* 45: 243–262.

RIESEBERG, L. H., AND GERBER, D. 1995. Hybridization in the Catalina mahogany: RAPD evidence. *Conservation Biology* 9: 199–203.

RIESEBERG, L. H., AND WENDEL, J. F. 1993. Introgression and its consequences in plants. *In* Hybrid Zones and the Evolutionary Process. *Edited by* R.G. Harrison. Oxford University Press, New York, New York pp. 70-109.

RODRIGO, A. G., M. KELLY-BORGES, P. R. BERGQUIST, AND P. L. BERGQUIST. 1993. A

randomisation test of the null hypothesis that two cladograms are sample estimates of a parametric phylogenetic tree. *New Zealand Journal of Botany* 31: 257-268.

ROKAS, A., N. KING, J. FINNERTY, AND S. B. CARROLL. 2003 a. Conflicting phylogenetic signals at the base of the metazoan tree. *Evolution and Development* 5: 346–359.

ROKAS, A., B. L. Williams, N. King, and S. B. Carroll. 2003 b. Genome-scale approaches to resolving incongruence in molecular phylogenies. *Nature* 425: 798-804.

RONALD, W. G. 1982. Intersectional hybridization of *Populus* sections, *Leuce-Aigeiros* and *Leuce-Tacamahaca*. *Silvae Genetica* 31: 94-99.

RONALD, W. G., LENZ, L. M., AND CUMMING, W. A. 1973 a. Biosystematics of the genus *Populus* L. I. Distribution and morphology of native Manitoba species and Variants. *Canadian Journal of Botany* 51: 2431-2442.

RONALD, W. G., LENZ, L. M., AND CUMMING, W. A. 1973 b. Biosystematics of the genus *Populus* L. II. Chemotaxonomy, phenology, fertility, and segregates of native Manitoba species and variants. *Canadian Journal of Botany* 51: 2443-2450.

ROOD, S. B., CAMPBELL, J. S., AND DESPINS, T. 1986. Natural poplar hybrids from southern Alberta. I. Continuous variation for foliar characteristics. *Canadian Journal of Botany* 64: 1382-1388

SATTA, Y., J. KLEIN, AND N. TAKAHATA. 2000. DNA archives and our nearest relative: the trichotomy problem revisited. *Molecular Phylogenetics and Evolution* 14: 259–275.

SCHREINER, E. J. 1974. *Populus* L. Poplar. *In* Seeds of woody plants in the United States. *Edited by* C.S. Schopmeyer. U.S. Department of Agriculture, Agriculture Handbook 450. Washington, DC. pp. 645-655.

SERVEDIO, M. R., AND KIRKPATRICK, M. 1997. The effects of gene flow on reinforcement. *Evolution* 51: 1764-1772.

SMITH, R. L. 1988. Phylogenetics of *Populus* L. (Salicaceae) based on restriction site fragment analysis of cpDNA. M.Sc. thesis. University of Wisconsin, Madison, WI., USA.

SMITH, R. L., AND K. J. SYTSMA. 1990. Evolution of *Populus nigra* (sect. *Aigeiros*): Introgressive hybridization and the chloroplast contribution of *Populus alba* (sect. *Populus*). *American Journal of Botany* 77: 1176-1187.

SOLTIS, P. S., D. E. SOLTIS, AND M. W. CHASE. 1999. Angiosperm phylogeny inferred from multiple genes as a tool for comparative biology. *Nature* 402: 402–404.

SORENSSON, C. T., AND BREWBAKER, J. L. 1994. Interspecies compatibility among 15 *Leucaena* species (Leguminosae: Mimosoideae) via artificial hybridizations. *American Journal of Botany* 81: 240-247.

STEBBINS, G. L. 1959. The role of hybridization in evolution. *Procceedings of the American Philosophical Society* 103: 231-251.

STEBBINS, G. L. 1950. Variation and evolution in plants. Columbia University Press, New York, New York.

STETTLER, R. F., H. D. BRADSHAW, JR., AND P. E. HEILMAN. 1996 a. Biology of *Populus* and its implications for management and conservation. NRC, Research Press, National Research Council of Canada, Ottawa, ON, Canada.

STETTLER, R. F., L. ZSUFFA, L., AND R. WU, 1996b. The role of hybridization in the genetic manipulation of *Populus*. pp. 87-112. *In* R. F. Stettler, H. D. Bradshaw, Jr., P. E. Heilman, and T. M. Hinckley [eds], Biology of *Populus* and its Implications for Management and Conservation. Part I, Chapter 4. NRC Research Press, National Research Council of Canada, Ottawa, ON, Canada.

SWOFFORD, D. L. 2001. PAUP*: Phylogenetic analysis using parsimony (* and other methods). Version 4.0b8 for Macintosh. Sinauer Associate, Sunderland, Massachusetts, USA.

TABERLET, P., GIELLY, L., PAUTOU, G., AND BOUVET, J. 1991. Universal primers for amplification of three non–coding regions of chloroplast DNA. *Plant Molecular Biology* 17: 1105–1109.

THOMPSON, J. D., D. G. HIGGINS, AND T. J. GIBSON. 1994. CLUSTAL W, improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position, specific gap penalties and matrix choice. *Nucleic Acids Research* 22: 4673– 4680.

TIFFIN, P., OLSON, M. S., AND MOYLE, L. C. 2001. Asymmetrical crossing barriers in angiosperms. *Proceedings of the Royal Society of London* 268: 861-867.

WANG, H., MCARTHUR, E. D., SANDERSON, S. C., GRAHAM, J. H., AND FREEMAN, D. C. 1997. Narrow hybrid zone between two subspecies of big sagebrush (*Artemisia tridentata*: Asteraceae). IV. Reciprocal transplant experiments. *Evolution* 51: 95-102.

WENDEL, J. F., AND J. J. DOYLE. 1998. DNA Sequencing. *In* D. E. Soltis, P. S. Soltis, and

J. J. Doyle [eds.], Molecular Systematics of Plants II, 265–296. Kluwer, Boston, Massachusetts, USA.

WHITHAM, T. G., K. D. FLOATE, G. D. MARTINSEN, E. M. DRIEBE, AND P. KEIM. 1996. Ecological and evolutionary implications of hybridization: *Populus*–herbivore interactions. *In* R. F. Stettler, H. D. Bradshaw, Jr., P.E. Heilman, and T. M. Hinckley [eds.], Biology of *Populus* and its implicationsfor management and conservation, 247–275. NRC Research Press, National Research Council of Canada, Ottawa, Ontario, Canada.

WHITHAM, T. G, MORROW, P. A., AND POTTS, B. M. 1994. Plant hybrid zones as center for biodiversity: the herbivore community of two endemic Tasmanian eucalypts. *Oecologia* 97: 481–90.

WIENS, J. J. 1998. Combining data sets with different phylogenetic histories. *Systematic Biology* 47: 568-581.

WOLFE, A. D., Q. Y. XIANG, AND S. R. KEPHART. 1998. Assessing hybridization in natural populations of *Penstemon* (Scrophulariaceae) using hypervariable intersimple sequence repeat (ISSR) bands. *Molecular Ecology* 77: 1107-1125.

WU, D. Y., WRIGHT, D. A., WETZEL, C., VOYTAS, D. F., AND RODERMEL, S. 1999. The immutans variegation locus of *Arabidopsis* defines a mitochondrial alternative oxidase homolog that functions during early chloroplast biogenesis. *Plant Cell* 11: 43-55.

YANG, Z., N. GOLDMAN, AND A. FRIDAY. 1994. Comparison of models for nucleotide substitution used in maximum-likelihood phylogenetic estimation. *Molecular and Biological Evolution* 11: 316–324.

ZIETKIEWICZ, E., A. RAFALSKI, AND D. LABUDA. 1994. Genome fingerprinting by simple sequence repeats (ISSR)-anchored polymerase chain reaction amplification. *Genomics* 20: 176-183.

ZSUFFA, L. 1975. A summary review of interspecific breeding in the genus *Populus*. *In* Proceedings of the 14th annual meeting of the Canadian Tree Improvement Association, part 2, 107–23. Canadian Forest Service, Ottawa, Ontario, Canada.