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A genetic study on subtropical *Anadenanthera colubrina* (Vell.) Brenan var. *cebil* (Griseb.) Altschul tree from Northwestern Argentina

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ABSTRACT
Anadenanthera colubrina (Vell.) Brenan var. *cebil* (Griseb.) Altschul is a tree species in South America important for its cultural, economic, and medicinal uses. In addition, it represents a trace in memory of the forests that have decreased over the years and for this reason it is not only interesting to study but also important to preserve the tree species for future generations. In this paper, we have characterized the genetic diversity of four populations. We collected seeds from four different sites: San Bernardo (B), El Cebilar (C), Metán (M), and El Gallinato (G) in Salta Province, North Argentina. We then compared the intergenic transcribed sequences of ribosomal DNA, a known genetic molecular marker. Our previous results, obtained through the morphological and genetic analysis of only four individuals (one for each zone), have showed that the individuals from B and M sites were more similar to each other as well as the individuals from G and C sites. In this paper, a larger number of individuals (25) were characterized and their phylogenetic relationships were computed. The results confirmed the previously found similarities.

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KEYWORDS
Biodiversity; botany; deforestation; conservation; phylogenetics

Introduction

In South America, a region that lies within the tropic, a series of humid-dry cycles occurred during the Pleistocene which altered the ecosystems. Several studies have been carried out, which have analysed the distribution of flora in relation to the climate during the last ice age, aimed at understanding the processes that produced vast areas of vegetation in South America (such as the Amazon rainforest and the grasslands of Patagonia) (Pennington et al. 2004). Glaciations and associated phenomena have had an important effect on the flora and fauna of South America, causing extinction and changes in the geographical distribution of species. They have produced the contraction of the forests during arid periods and their expansion during wet periods (Van der Hammen 1974). Climate changes during the Quaternary ages have caused fluctuations of population size (Hewitt 2000).

Anadenanthera colubrina (Vell.) Brenan var. *cebil* (Griseb.) Altschul belongs to the Fabaceae family, Mimosoideae subfamily (http://www.kew.org/science-conservation/research-data/resources/legumes-of-the-world), and has a wide distribution in Brazil, Paraguay, Perù, Bolivia, and Argentina (http://floradobrasil.jbrj.gov.br/jabot/floradobrasil/FB18071). In Argentina, it is located in the northern provinces. Today, *A. colubrina* var. *cebil* is intermittently distributed in the seasonally dry tropical forests (Särkinen et al. 2011). This species was categorized as one of "least concern" with a low risk of extinction by IUCN (http://www.iucnredlist.org), although del Valle Perea (www.atlas.catamarca.gov.ar) considered it a vulnerable species in Catamarca, with many populations at risk due to its continuous exploitation. A large area of this forest has destroyed, due to the improvement of agricultural soils, and so the number of *A. colubrina* var. *cebil* has decreased in recent decades representing an example of species fragmentation after deforestation (Barrandeguy et al. 2014).

The further importance of this tree species is due to its economic and medicinal applications in South America. In fact, the wood contains tannins, useful in processing animal hides; it is hard and termite resistant and for this reason it is used in furniture. In popular medicine, it is used to treat respiratory diseases (Carod-Artal & Vásquez-Cabrera 2007).

Intergenic transcribed spacers (ITS) of ribosomal DNA (rDNA) represents genetic molecular markers known to be useful in phylogenetics used in detecting both interspecific and intraspecific variability, as they contain conserved and variable regions (Simon & Weiß 2008). For example, in fungi, the use of ITS marker has allowed a better understanding of intraspecific variability (Schoch et al. 2012).

As demonstrated in a previous study (de Viana et al. 2014), the analysis of the morphological traits in the fruits and seeds of *A. colubrina* var. *cebil* showed similarities among trees from different regions of the same province of Salta (north Argentina): Metán, El Cebilar, San Bernardo, and El Gallinato. The authors analysed the ITS region of fresh seeds, from four trees of *A. colubrina* var. *cebil* (one from each site) collected in Salta Province, showing a wide variability among individuals at both morphologic and genetic levels.

The aim of this work was to deepen the previous study by including a larger number of individuals (25) collected from different locations in Salta Province (Figure 1) and conserved in the Germplasm Bank of Native Species (BGEN-INEAH) of the National University of Salta through

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Supplemental data for this article can be accessed here.

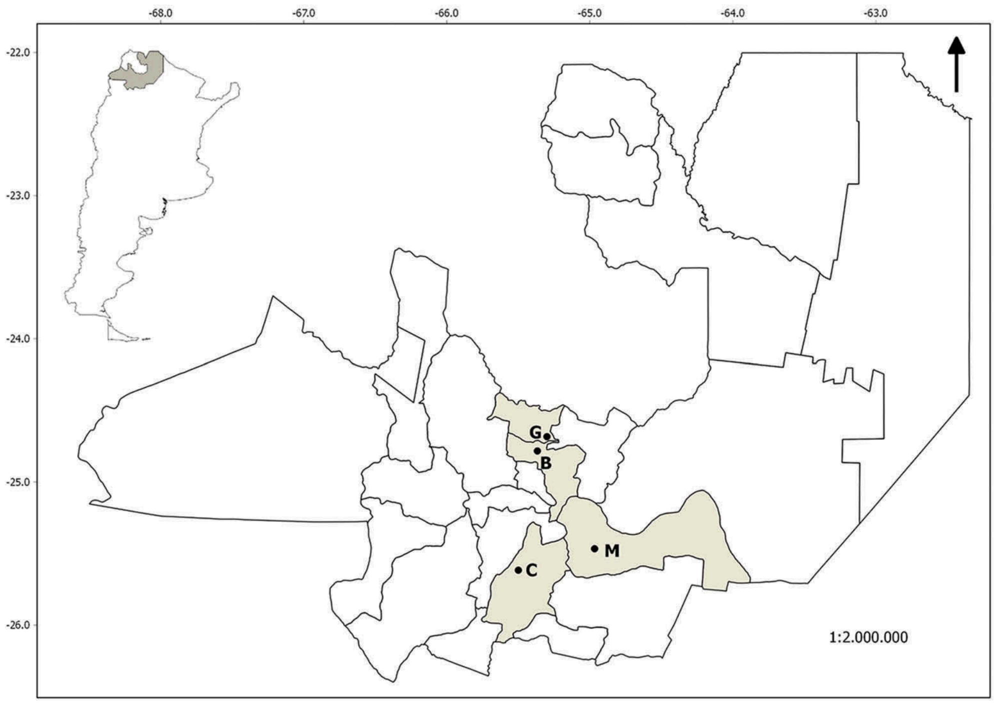

Figure 1. Map of Argentina and Salta Province. The four sites of sampling, San Bernardo (B); El Cebilar (C); El Gallinato (G); and Metán (M).

the use of the ITS-rDNA marker. Study on the genetic diversity of *A. colubrina* var. *cebil* may improve our knowledge about plant genetic characteristics and indicate a tool for propagation and conservation policies.

Materials and methods

Sampling and DNA extraction from plant seeds

Seeds of *A. colubrina* var. *cebil* were collected from four regions in Salta Province including San Bernardo (B), El Cebilar (C), Metán (M), and El Gallinato (G), which are located within about 100 km of each other, as shown in Figure 1. We collected 100 fruits per individual tree, for a total of 10 individuals per population. In the laboratory, the fruits were opened manually and most of the seeds were kept for long-term *ex situ* conservation at the germplasm bank in Salta Province. The seeds used in this study, obtained from different trees, were sterilized and shredded; then the genomic DNA was extracted as previously described (de Viana et al. 2014). Environmental and geographical data of the collecting sites are reported in Table 1.

In vitro amplification (PCR) of ITS regions and analysis of the nucleotide sequences

PCR was carried out as previously described (de Viana et al. 2014). The size of the specific isolated region was 450bp. The 25 amplicons nucleotide composition was determined by the MWG sequence service (Eurofin-operon) and published in GeneBank. The relative accession numbers are included in Table S1.

Nucleotide alignment, phylogenetic and polymorphic analyses

Multiple alignments of the ITS-rDNA sequences were performed with Clustal W2 software (http://www.expasy.org). Shading of the multiple alignment sequences were obtained by Boxshade 3.21 program (http://www.ch.embnet.org/software/BOX_form.html).

The phylogenetic tree was achieved by MEGA6 (Tamura et al. 2013), using the neighbor-joining method (with 500 bootstrap replications). The genetic distances were computed using the Maximum Composite Likelihood Method (Tamura et al. 2004) and are in the units of the number of base substitutions per site. Codon positions included were 1st+2nd+3rd+noncoding. *Cojoba undulatomarginata* L.Rico (Accession number EF638187) and *Calliandra physocalyx* H.M. Hern. & M. Sousa (Accession number JX870727) were utilized as outgroups because of their belonging to the same family (Fabaceae) of *A. colubrina* but are appropriately distant.

The DnaSP (Version 5.0) (http://www.ub.edu/dnasp) platform was used to sort the number of variable sites.

Table 1. Geographical and climate data of the Argentina collecting sites (period 1934–1990) (http://anterior.inta.gob.ar/prorenoa/info/resultados/Precip_NOA/base_precipitaciones_noa.asp).

Recording station	Collecting sites	Geographical coordinates Latitude–longitude	Altitude (MASL)	Average annual rainfall (mm)	Average annual temperature (°C)	N° individuals for each site
Salta	San Bernardo	24°47′S–65°22′W	1410	695 ± 170	16	5
Mojotoro	El Gallinato	24°41′S–65°18′W	1190	607 ± 187	16	8
Talapampa	El Cebilar	25°37′S–65°30′W	1432	376 ± 73	16	8
Metán	Metán	25°38′S–64°58′W	884	902 ± 259	18	8

MASL = meters above sea level.

Results

Phylogenetic and polymorphic analysis

The phylogenetic relationship between 25 different individuals of *A. colubrina* var. *cebil*, collected in four regions of Salta Province, San Bernardo (B), El Cebilar (C), Metán (M), and El Gallinato (G), was investigated through the ITS-rDNA region sequences. In this study, the sequences previously identified as AcC, AcG, AcM, AcB (de Viana et al. 2014) were also included. The aligned nucleotidic sequences are shown in Table S2 and the related nucleotidic differences are shaded in Table S3. The phylogenetic tree constructed by using the 29 sequences is shown in Figure 2. All of the individuals were sorted into two main groups, which branch out separately, containing 16 and 13 sequences, respectively. The two outgroup sequences of the plants *C. undulatomarginata* and *C. physocalyx* have clustered in a third group, as expected. In this phylogenetic tree, all individuals are divided into two branches: one giving rise to the AcC/AcG group and another giving rise to the AcM/AcB group. Polymorphic analysis was sorted using DnaSP software. A total of 225 variable sites were detected out of 477 total nucleotides. Genetic distance among the four groups of populations, found by using MEGA 6 software, showed the following results: the distance between M and G populations was 0.191, between M and C was 0.203, and between M and B was 0.07; the distance between C and G populations was 0.08, and between C and B was 0.174; finally, the distance between B and G populations was 0.163 (see Table S4).

Discussion

The choice of ITS-rDNA as a marker capable of establishing the genetic variability of *A. colubrina* var. *cebil* is related to the variability of these nonfunctional sequences on which a low-selective pressure acts. Thus, the ITS-rDNA marker is widely used to study interspecific and intraspecific variations in plants such as Asteraceae and Cyperaceae (Baldwin et al. 1995; Kollipara et al. 1997). In

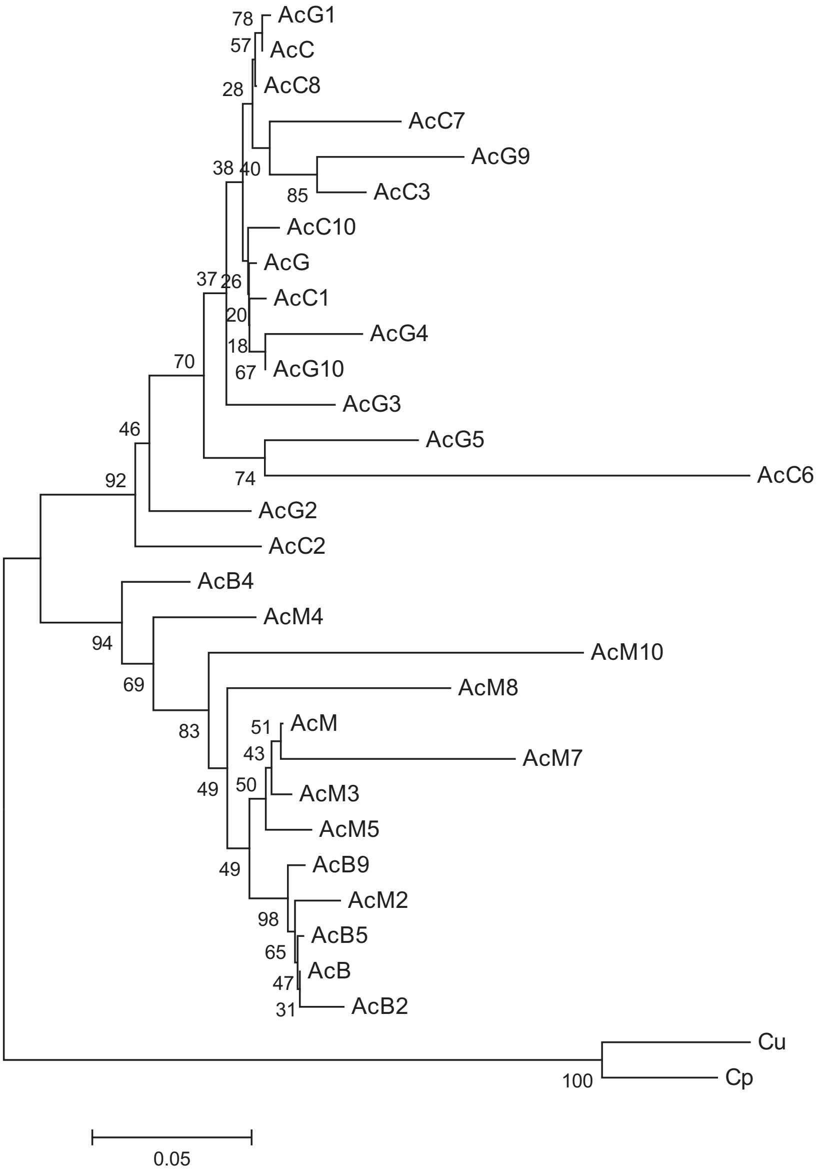

Figure 2. Phylogenetic tree of *Anadenanthera colubrina* var. *cebil* populations. The four sites of sampling were B = San Bernardo; C = El Cebilar; G = El Gallinato; M = Metán. *Cojoba undulatomarginata* (Cu) and *Calliandra physocalyx* (Cp), which are plants of the same family (Fabaceae) as *A. colubrina*, used as outgroups. The tree is drawn to scale, with branch lengths in the same units as those of the genetic distances. The genetic distances are in units of the number of base substitutions per site. The high bootstrap values indicate the accuracy of the data.

this paper, the sequence analysis of the 29 samples showed that the Argentine population of *A. colubrina* var. *cebil* possesses a genetic variability within species, as deduced by the phylogenetic tree (Figure 2). In particular, the cluster analysis displayed a high similarity between individuals collected from the regions of San Bernardo and Metán, and between individuals collected from the regions of El Cebilar and El Gallinato (Figure 1). The genetic distance, found between the four analysed populations, clearly showed the same results. In fact, individuals from G/C and B/M sites belonged to the same cluster in the phylogenetic tree. The genetic distance confirmed these findings. In fact, the distance between B and M (corresponding to a value equal to 0.07) was lower than that between B and C/G (0.203 and 0.163, respectively). Analogously, the distance between G and C was 0.08 (see Table S4).

These results are in agreement with the previous study (de Viana et al. 2014) in which the comparison between the sequences of four individuals, one per each region, revealed high identity (98.1%) between B and M, as well as between C and G (97.9%). It could be conceivable to hypothesize that all these individuals belong to a single large population of *A. colubrina* var. *cebil* with a distribution area that extends from G/B (north) and C/M (south). The environmental conditions and geographical data of the seeds-collecting sites, relating to the period 1934–1990, are reported in Table 1.

The distribution of different individuals may suggest progressive divergence, due to isolation, as in the study on molecular markers in Anacardiaceae reported by Caetano et al. (2008). Alternatively, but not exclusively, populations of *A. colubrina* var. *cebil* from Salta Province had genetic diversity in ITS-rDNA sequences, simply as a consequence of genetic drift.

Our results are in agreement with those reported by Barrandeguy et al. (2012) and Garcia et al. (2014). They found genetic variability and high levels of polymorphism in natural populations of *A. colubrina* var. *cebil* collected from northern provinces of Argentina, other than Salta. In addition, they used chloroplast microsatellite (cpSSR) plus nuclear SSRs loci (nSSRs), other than ITS marker.

As regards the high grade of intraspecific polymorphism detected in our study, it is known that ITS polymorphism within individuals is quite common (Baldwin et al. 1995; Rosselló et al. 2006).

As trees have a crucial role in the social, ecological, and economic life of the Earth, covering 30% of its surface, the application of genetics to plant diversity may determine improvements in knowledge about conservation and forests management (Rao & Hodgkin 2002).

The assessment of intraspecific genetic variability of *A. colubrina* var. *cebil* is important to design appropriate conservation policies. Further studies are needed to assess the levels of variability of a larger population which also includes neighbouring countries of subtropical South America.

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Disclosure statement

No potential conflict of interest was reported by the authors.

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Table S1 Accession numbers of the *Anadanthera colubrina* samples

Samples	**Accession numbers**
AcM	JQ910930
AcM2	LN827632
AcM3	LN827633
AcM4	LN827634
AcM5	LN827635
AcM7	LN827636
AcM8	LN827637
AcM10	LN827638
AcC	HF564642
AcC1	LN827646
AcC2	LN827647
AcC3	LN827648
AcC6	LN827649
AcC7	LN827650
AcC8	LN827651
AcC10	LN827652
AcB	HF564640
AcB2	LN827653
AcB4	LN827654
AcB5	LN827655
AcB9	LN827656
AcG	HF564641
AcG1	LN827639

AcG2	LN827640
AcG3	LN827641
AcG4	LN827642
AcG5	LN827643
AcG9	LN827644
AcG10	LN827645

Table S2 Clustal W2 alignment

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ACG-HF564641.1      -------TGAAAGCCT-CCCAAGCCCGGGCACAACA--CAAACCCCGGTACCTCAGGTGC 50
ACC1                -------TGAAAGCCT-CCCAAGCCCGGGCACAACA--CAAACCCCGGTACCTCAGGTGC 50
ACC10               -------TGAAAGCCT-CCCAAGCCCGGGCACAACG--CCAACCCCGGCACCTCAGGTGC 50
ACG1                -------TGAAAGCCT-CCCAAGCCCGGGTACAACA--CAAACGCCGGTACCTCAGGTGC 50
ACC-HF564642.1      -------TGAAAGCCT-CCCAAGCCCGGGTACAACA--CAAACGCCGGTACCTCAGGTGC 50
ACG9                -------TGAAAGCCT-CCCAAGCCCGGGCACAACA--CAAACGCCGGTACCTCAGGTGC 50
ACC3                -------TGAAAGCCT-CCCAAGCCCGGGTACAACA--CAAACGCCGGTACCTCAGGTGC 50
ACC7                -------TGAAAGCCT-CCCAAGCCCGGGTACGACG--CAAACGCTGGTACCTCAGGTGC 50
ACC8                -------TGAAAGCCT-CCCAAGCCCGGGTACAACA--CAAACGCCGGTACCTCAGGTGC 50
ACG4                -------TGAAAGCCT-CCCAAGCCCGGGCACAACA--CAAACCCCGGTACCTCAGGTGC 50
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ACC6                -------TGAAAGCCT-CCCGAGCGCGGGCACAACG--CCTACCCCGGCACCTTAGGTGC 50
ACM-JQ910930.1      -------TGAAAGCCT-CCCAAGCCCGGGCACAACA--CAAACCCTGGTGCCTCAGGCAC 50
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ACM5                -------TGAAAGCCT-CCCAAGCCCGGGCACAACA--CAAACCCTGGTGCCTCAGGCAC 50
ACB-HF564640.1      -------TGAAAGCCT-CCCAAGCCCGGGCACAACA--CAAACCCTGGTGCCTCAGGCAC 50
ACB2                -------TGAAAGCCT-CCCAAGCCCGGGCACAACA--CAAACCCTGGTGCCTCAGGCAC 50
ACB5                -------TGAAAGCCT-CCCAAGCCCGGGCACAACA--CAAACCCTGGTGCCTCAGGCAC 50
ACM2                -------TGAAAGCCT-CCCAAGCCCGGGCACAACA--CAAACCCTGGTGCCTCAGGCAC 50
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ACM10               -------TGAAAGCCT-CCCAAGCCAGGGCACAACA--CAAACCCTGGTGCCTCAGGCAC 50
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AcB4                -------TGAAAGCCT-CCCAAGCCCGGGCACAACA--CAAACCCTGGTACCTCAGGTAC 50
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CP                  ----GG-----GGCGCGCCCGAGGCC-CCCCCGACAACCAAACCCCGGCGCCACAGGCGC 50
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ACG-HF564641.1      CAAGGAACTGCAACAGAGCGAG-AGCGCCTTACGCCGATTGCACCCGAAATGACTCTCAA 109
ACC1                CAAGGAACTGCAACAGAGCGAG-AGTGCCTTACGTCGATTGCACCCGAAATGACTCTCAA 109
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ACG3                CAAGGAACTGCAACAGAGCGAG-TGCGCCTTACGCCGATTGCACCCGAAATGACTCTCAA 109
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ACC6                CAAGCAACTGCAACAGAGGGAG-AGCGCCTCTTGCCGATTGCACCCAAATGCACTCTTAA 109
ACM-JQ910930.1      CAAGGAACTGCAACAGAGCGAG-TGCGCCTCATGCCGATTGCACCCAAAACGACTCTCGG 109
ACM3                CAAGGAACTGCAACAGAGCGAG-TGCGCCTCATGCCGATTGCACCCAAAACGACTCTCGG 109
ACM5                CAAGGAACTGCAACAGAGCGAG-TGCGCCTCATGCCGATTGCACCCAAAACGACTCTCGG 109
ACB-HF564640.1      CAAGGAACTGCAACAGAGCGAG-TGCGCCTCATGCCGATTGCACCCAAAACGACTCTCGG 109
ACB2                CAAGGAACTGCAACAGAGCGAG-TGCGCCTCATGCCGATTGCACCCAAAACGACTCTCGG 109
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ACM2                CAAGGAACTGCAACAGAGCGAG-TGCGCCTCATGCCGATTGCACCCAAAACGACTCTCGG 109
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CP                  CAAGGAACAGCAAAGAAGCAAGGCACGCATGATGCCATTTGCATCCGAAACGACTCTCGG 110
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ACG1                CAACGGATATCTCAGCTCTCGCATCGATGAAGAACGTAGTGAAATGCAATGCTTGGTGTG 169
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ACC3                TAACGGATATCTCAGCTCTCGCATCAATGAAGAACGTAGTGAAATGCGATGCTTGGTGTG 169
ACC7                CAACGGATATCTCAGCTCTCGCATCGATGAAGAACGTAGGGAAATGCAATACTTGGTGTG 169
ACC8                CAACGGATATCTCAGCTCTCGCATCGATGAAGAACGTAGTGAAATGCAATGCTTGGTGTG 169
ACG4                CAACGGATATCTCAGCTCTCGCATCGATGAAGAACGTAGTGAAATGCAATGCTTGGTGTG 169
ACG10               CAACGGATATCTCAGCTCTCGCATCGATGAAGAACGTAGTGAAATGCAATGCTTGGTGTG 169
ACG3                CAACGGATATCTCAGCTCTCGCATCAATGAAGAACGTAGTGAAATGCAATGCTTGGTGTG 169
ACG5                CAACGGATATCTCAGCTCTCGCATCGATGAAGAACGTAGTGGAATGCGATGCTTGGTGTG 169
ACG2                CAACGGATATCTCAGCTCTCGCATCGATGAAGAACATAGCGAAATGCGATGCTTAGTGTG 169
ACC2                CAACGGATATCTCAGCTCTCGCATCGATGAAGAACGTAGCGAAATGCAATGCTTAGTGTG 169
ACC6                CAACGGATATCTCACTTCTGGCATCGATGAAGAACGTCGCGAAATGCAATGCTTGGTGTG 169
ACM-JQ910930.1      CAACGGATATCTCGGCTCTCGCATCGATGAAGAACATAGCGAAATGCGATGCTTGGTGTG 169
ACM3                CAACGGATATCTCGGCTCTCGCATCGATGAAGAACATAGCGAAATGCGATGCTTGGTGTG 169
ACM5                CAACGGATATCTCGGCTCTCGCATCGATGAAGAACATAGCGAAATGCGATGCTTGGTGTG 169
ACB-HF564640.1      CAACGGATATCTCGACTCTCGCATCGATGAAGAACATAGCGAAATGCGATGCTTAGTGTG 169
ACB2                CAACGGATATCTCGACTCTTGGATCGATGAAGAACATAGCGAAATGCGATGCTTAGTGTG 169
ACB5                CAACGGATATCTCGACTCTCGCATCGATGAAGAACATAGCGAAATGCGATGCTTAGTGTG 169
ACM2                CAACGGATATCTCGACTCTCGCATCGATGAAGAACATAGCGAAATGCGATGCTTAGTGTG 169
ACB9                CAACGGATATCTCGACTCTCGCATCGATGAAGAACATAGCGAAATGCGATGCTTGGTGTG 169
ACM7                TACCGGATATCTCGGCTCTCGCATCGATGAAGAACATAGCGAAATGCGATGCTTGGTGTG 169
ACM8                CAACGGATATCTCGGCTCTCGCATCGATGAAGAACATAGCGAAATGTGATGCTTGGTGTG 169
ACM10               CAACGTATATCTTAGCTCTCATATCTAAAAAGAACAGAGCGAAATATGATACTTGGTGTG 169
AcM4                CAAAGGATATCTCGGCTCTCGCATCGATGAAGAACATAGCGAAATGCAATGCTTGGTGTG 169
AcB4                CAACGGATATCTCGACTCTCGCATCGATGAAGAACATAGCGAAATGCAATGCTTGGTGTG 169
CU                  CAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTG 177
CP                  CAACGGATATCTCGGCTCTCGCATCGATGAAGAACGTAGCGAAATGCGATACTTGGTGTG 170
                     *..* ****** .  *** . *** *:. *****. .* *.***. .**.***.*****
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ACG-HF564641.1      AATTGCAGAATCCTGTGAACTATCGAGTCTTTGAATGTAAGTTGCG-CCAAAGCTATTAG 227
ACC1                AATTGCAGAATCCTGTGAACTATCGAGTCTTTGAATGTAAGTTGCGCCCAAAGCTATTAG 229
ACC10               AATTGCAGAATCCTGTGAACTATCGAGTCTTTGAATGTAAGTTGCGCCCAAAGCTATTAG 229
ACG1                AATTGCAGAATCCTGTGAACTATTGAGTCTTTGAATGTAAGTTGCGCCCAAAGCTATTAG 229
ACC-HF564642.1      AATTGCAGAATCCTGTGAACTATTGAGTCTTTGAATGTAAGTTGCG-CCAAAGCTATTAG 228
ACG9                AATTGCAGAATCCCGTGAACCATGGAGTCTTTGAATGTAAGTTACGCCCGAAGCTATTAG 229
ACC3                AATTGCAGAATCCCGTGAACCATTGAGTCTTTGAATGCAAGTTACGCCCAAAGCTATTAG 229
ACC7                AATTGCAAAATCCCGTGAACTATCGAGTCTTTGAATGCAAGTTGCGCCCAAAGCCATTAG 229
ACC8                AATTGCAGAATCCTGTGAACTATCGAGTCTTTGAATGTAAGTTGCGCCCAAAGCTATTAG 229
ACG4                AATTGCAGAATCCTGTGAACTATCGAGTCTTTGAATGTAAGTTGCGCCCAAAGCTATTAG 229
ACG10               AATTGCAGAATCCTGTGAACTATCGAGTCTTTGAATGTAAGTTGCGCCCAAAGCTATTAG 229
ACG3                AATTGCAGAATCCTGTGAACTATCGAGTCTTTGAATGTAAGTTGCGCCCAAAGCTATTAG 229
ACG5                AATTGCAGAATCCTGTGAACTATTGAGTCTTTGAATGTAAGTTATGCCCCATGCTATTAG 229
ACG2                AATTGCAGAATCCTGTGAACTATTGAGTCTTTGAACGCAAGTTGCGCCCAAAGCTATTAG 229
ACC2                AATTGCAGAATCCCGTGAACCATTGAGTCCTTGAACGCAAGTTGTG-CCAAAGCCATTAG 228
ACC6                AATTGCGGAATCCTGAGAACTATTGAGTCTTTGAAGGGAACTTGTGCCCAAATTGATTAG 229
ACM-JQ910930.1      AATTGCAGAATCCCGTGAACCATTGAGTCCTTGAACGCAAGTTGTGCCCGAAGCCATTAG 229
ACM3                AATTGCAGAATCCCGTGAACCATTGAGTCCTTGAACGCAAGTTGTGCCCGAAGCCATTAG 229
ACM5                AATTGCAGAATTCCGGGAACCATTGAGTCCTTGAACGCAAGTTGTGCCCGAAGCCATTAG 229
ACB-HF564640.1      AATTGCAGAATCCCGTGAACCATTGAGTCCTTGAACGCAAGTTGTGCCCAAAGCCATTAG 229
ACB2                AATTGCAGAATCCCGTGAACCATTTAGTCCTTGAACGCAAGTTGTGCCCAAAGCCATTAG 229
ACB5                AATTGCAGAATCCCGTGAACCATTGAGTCCTTGAACGCAAGTTGTGCCCAAAGCCATTAG 229
ACM2                AATTGCAGAATCCCGTGAACCATTGAGTCCTTGAACGCAAGTTGTGCCCAAAGCCATTAG 229
ACB9                AATTGCAGAATCCCGTGAACCATTGAGTCCTTGAACGCAAGTTGTGCCCAAAGCCATTAG 229
ACM7                AATTGCAGAATCCCGTGAACCATTGAGTCCTTGAACGCAAGTTGTGCCCGAAGCCATTAG 229
ACM8                ACTTGCAGAATCCTGTGAACCATCGAGTCCTTGAACGCAAGTTGTGCCCGAAGTCATTAG 229
ACM10               AAGTGGAGAATCCCGTGAACCATGGAGTCTTTGAATGCAAGTTACGCCCGAAGCCATTAG 229
AcM4                AATTGCAGAATCCTGTTAACCATTGAGTCCTTGAACGGAAGTTGTGCCCGAAGCCATTAG 229
AcB4                AATTGCAGAATCCCGTGAACCATCGAGTCTTTGAACGTAAGTTGCGCCCAAAGCTATTAG 229
CU                  AATTGCAGAATCCCGTGAACCATCGAGTCTTTGAACGCAAGTTGCGCCCGAAGCCTTTAG 237
CP                  AATTGCAGAATCCCGTGAACCATCGAGTCTTTGAACGCAAGTTGCGCCCGAAGCCTTTAG 230
                    *. ** ..*** * *  *** **  **** ***** * ** **. * ** *:   :****
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ACG-HF564641.1      GCCGAGGGCACGCCTGCCTGGGCGTCACGCAACGTCGCCGCCACCCCACGCG--GGGCAC 285
ACC1                GCCGAGGGCACGCCTGCCTGGGCGTCACGCAACGTCGCCGCCACCCCACGCG--GGGCAC 287
ACC10               GCCGAGGGCACGCCTGCCTGGGCGTCACGCAACGTCGCCGCCACCCCACGCG--GGGCAC 287
ACG1                GCCGAGGGCACGCCTGCCTGGGCGTCACGCAACGTCGCCGCCACCCCACGCG--GGGCAC 287
ACC-HF564642.1      GCCGAGGGCACGCCTGCCTGGGCGTCACGCAACGTCGCCGCCACCCCACGCG--GGGCAC 286
ACG9                GCTGAGGGCACGCCTGCCTAGGCGTCACGTAACATCGCCGCCACCCCACGCG--GGGCAC 287
ACC3                GCCGAGGGCACGCCTGCCTGGGCGTCACGTAACGTCGCCGCCACCCCACGCG--GGGCAC 287
ACC7                GCCGAGGGCACGCCTGCTTGGGCGTCACGCAAGGTCGCCGCCACCCCACGCG--GGGCAC 287
ACC8                GCCGAGGGCACGCCTGCCTGGGCGTCACGCAACGTCGCCGCCACCCCACGCG--GGGCAC 287
ACG4                GCCGAGGGCACGCCTGCCATGGCGACACGCAACGTCGCCGCCACCCCACGCG--GAGCAC 287
ACG10               GCCGAGGGCACGCCTGCCTGGGCGTCACGCAACGTCGCCGCCACCCCACGCG--GGGCAC 287
ACG3                GCCGAGGGCACGCCTGCCTGGGCATCACGCAACGTTGCCGCCACCCCACGCG--GGGCCC 287
ACG5                GCCGAGGGCACGCCTGCCTGGGCGTCAGGCAACGTCGCCGCCACCTCACGCG--GGGCGC 287
ACG2                GCCGAGGGCACGCCTGCCCGGGCATCACGCAACGTTGCCGCCGCCTCACGCG--GGGCAC 287
ACC2                GCCGAGGGCACGCCTGCCCGGGCATCACGCAACGTTGCCGCCGCCTCACGCG--AGGCGC 286
ACC6                GCGGAGGGCCCGCCTGCATAGGCCTGAGGCAACGCCGCCGCGCCCTCACGCG--GGGCGC 287
ACM-JQ910930.1      GCCGAGGGCACGCCTGCCCGGGCATCACGCAACGTCGCCGCTACCTCACGCG--GGGCGC 287
ACM3                GCCGAGGGCACGCCTGCCCGGGCATCACGCAACGTCGCCGCCGCCTCACGCG--GGGCGC 287
ACM5                GCCGAGGGCACGCGTGCCCGGGCATCACGCAACGTCGCCGCGACCTCACGCG--GGGCGC 287
ACB-HF564640.1      GCCGAGGGCACGCCTGCCCGGGCATCACGCAACGTTGCCGCCGCCTCACGCG--GGGCGC 287
ACB2                GCCGAGGGCACGCTTGCCCGGGCATCACGCAACGTTGCCGCCGCCTCACGCG--GGGCGC 287
ACB5                GCCGAGGGCACGCCTGCCCGGGCATCACGCAACGTTGCCGCCGCCTCACGCG--GGGCGC 287
ACM2                GCCGAGGGCACGCCTGCCCGGGCATCACGCAACGTTGCCGCCGCCTCACGCG--GGGCGC 287
ACB9                GCCGAGGGCACGCCTGCCCGGGCATCACGCAACGTTGCCGCCGCCTCACGCG--GGGCGC 287
ACM7                GCCAAGGACACGCCTGCTTGGGCATCACGCATCGTCGCGCCTACCTCACGCG--GGGCGC 287
ACM8                GCCGAGGGCACGCCTGCCCTTGCGTCACGTATCGTCGCCGCGCCCTCAAGCG--GGGCGC 287
ACM10               GCTGAGGGCACCCCTGCCTGGGCGTCACGTAACGTCGCGGCTACCTGACGCG--GGGCGC 287
AcM4                GCCGAGGGCACGCCTGCCCGGGCCTCACGGAACGTCGCCGCCACCCCACGCG--GGGCGG 287
AcB4                GCCGAGGGCACGCCTGCCTGGGCATCACGCAACGTCGCCGCCACCTCACGCG--GGGCGC 287
CU                  GCCGAGGGCACGTCTGCCTGGGTGTCACGCAACGTCGCCAACGCCCGATCCTCTGGGCGT 297
CP                  GCCGAGGGCACGTCTGCCTGGGTGTCACGCAGCGTCGCCACCGCCCGATCATGCGGGCTT 290
                    ** .***.*.*   ***    *  : * * *  .  **  .  **  *  .   ..**
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ACG-HF564641.1      GGCGAATGATGGCCTCCCGAGGCCTCTATCCCCCAGCTGGCCGAAATAACTGCCTAGCGC 345
ACC1                GGCGAATGATGGCCTCCCGAGGCCTCTATCCCCCAGCTGGCCGAAATAACTGCCTAGCGC 347
ACC10               GGCGAATGATGGCCTCCCGAGGCCTCTATCCCCCAGCTGGCCGAAATAACTGCCTAGCGC 347
ACG1                GGCGAATGATGGCCTCCCGAGGCCTCTATCCCCCAGCTGGCCGAAATAACTGCCTAGCGC 347
ACC-HF564642.1      GGCGAATGATGGCCTCCCGAGGCCTCTATCCCCCAGCTGGCCGAAATAACTGCCTAGCGC 346
ACG9                GGTGAATGATGGCCTCCCGAGGCCTCTGTCCCCCAGCTGGCTGAAATGACGGCCTAGCGC 347
ACC3                GGCGAATGATGGCCTCCCGAGGCCTCTATCCCCCAGCTGGCCGAAATAACTGCCTAGCGC 347
ACC7                GGCGAATGATGGCCTCCCGAGGCCTCTATCCCCCAGCTGGCCGAAATAACTGCCTAGCGC 347
ACC8                GGCGAATGATGGCCTCCCGAGGCCTCTATCCCCCAGCTGGCCGAAATAACTGCCTAGCGC 347
ACG4                GGCGAATGATGGCCTCCCAGAGGCTCTATCCCCTAACTGGCCGAAATAACTGCCTAGCGC 347
ACG10               GGCGAATGATGGCCTCCCAAGGCCTCTATCCCCCAACTGGCCGAAATAACTGCCTAGCGC 347
ACG3                GGTGAATGATGGCCTCCCGAGGCCTCCATCCCCCAGCTGGCCGAAATAACTGCCTCGCGC 347
ACG5                CGCAAATGATGGCCTCCCCGAGGCTCTATCCCCCAGCTGGACGAAATCACTGCCTAGCGC 347
ACG2                GGCGAATGATGGCCTCCCGAGGCCTCTATCCCCCGGCTGGCCGAAATAACTGCCTCGCGC 347
ACC2                GGCGAATGATGGCCTCCCGGGGCCTCCATCCCCTAGCTAGCCGAAATGACGGCCTAGCGC 346
ACC6                CGCAGATGATGGCGTCCCGAGGGCTCTATCCCCCGGCTAGCGGAAATGACTCCCTAGCGT 347
ACM-JQ910930.1      GGTAGATGATGGCCTCCTAGGGGCTCCATCCCCTGGCTAGCTGAAATGATGGCCTCGCGC 347
ACM3                GGTAGATGATGGCCTCCTAGGGGCTCCATCCCCTGGCTAGCTGAAATGATGGCCTTACGC 347
ACM5                GGTAGATGATGGCCTCCTAGGGGCTCCATCCCCTGGCTAGCTGAAATGATGGCCTAGCGC 347
ACB-HF564640.1      GGTAGATGATGGCCTCCCAGGGGCTCCATCCCCTGGCTAGCTGAAATGATGGCCTCGCGC 347
ACB2                GGTAGATGATGGCCTCCCAGGGGCTCCATCCCCTGGCTAGGTGAAATGATGGCCTCGCGC 347
ACB5                GGTAGATGATGGCCTCCCAGGGGCTCCATCCCCTGGCTAGCTGAAATGATGGCCTCACGC 347
ACM2                GATAGATGATGACCTCCCAGGGGCTCGATCCCCTGGCTAGCTGAAATGATGGCCTCGCGC 347
ACB9                GGTAGATGATGGCCTCCCAGGGGCTCCATCCCCTGGCTAGCTGAAATGATGGCCTCGCGC 347
ACM7                GGTAGAAGATGGCTTCTTAGGGCCTCCATCCCCTGGCTACTTGAATTGATGCCCTCCCCC 347
ACM8                GGTAGATGATGGCCTCTTTGGGGCTCCATCCTCCGGCTAGCCGAAATGATGGCCTAGTGC 347
ACM10               GGTTGATGATGGCCTCCTAGGGGCTCCATCCCCTGGCTAGCTGAAATGTTGGCCTCGCGC 347
AcM4                GGTGGATGATGGCCTCCCAGGGGCTCCATCCCCCGGCTGGCTGAAATGATGGCCTCGCGC 347
AcB4                GGTAGATGATGGCCTCCCAGGGGCTCCATCCCCAGGCTGGCTGAAATAATGGCCTAGCGC 347
CU                  GGCGGATGATGGCCTCCCGGGAGCCTCGCCTCCCGGCTGGCCGAAAAAGGGGCCCGACGT 357
CP                  GGCGGATGATGGCCTCCCGGGAGCGTCGCCTCCCGGCTGGCCGAAATAGGGGCCCAACGT 350
                     .  .*:****.* **   ... *   . *  * ..**.   ***::     **
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ACG-HF564641.1          GATGGCGGCAATGATCCACGGGGACGTTCGAGACCGAACGTGATCG-CACAATCCCATGG 404
ACC1                    GATGGCGGCCATGATCCACGGGGACGTTCGAGACCGAACGTGATCG-CACAATCCCATGG 406
ACC10                   GATGGCGGCCATGATCCACGGGGACGTTCGAGACCGAACGTGATCG-CACAATCCCATGG 406
ACG1                    GATGGCGGCCATGATCCACGGGGACGTTCGAGACCGAACGTGATCG-CACAATCCCATGG 406
ACC-HF564642.1          GATGGCGGCCATGATCCACGGGGACGTTCGAGACCGAACGTGATCG-CACAATCCCATGG 405
ACG9                    GACGGTGGCCACGATCCACGAGGACGCTTGAGACCGAACGTGATCG-CACAATCCCATGG 406
ACC3                    GATGGCGGCCATGATCCACGGGGACGTTCGAGACCGAACGTGATCG-CACAATCCCATGG 406
ACC7                    AATGGCGGCCATGATCCACGGAGAAGTTCGAGACCGAACGTGATCG-CACAATCCCATGG 406
ACC8                    GATGGCGGCCATGATCCACGGGGACGTTCGAGACCGAACGTGATCG-CACAATCCCATGG 406
ACG4                    GATGGCGGCCATGATCCACGTGGACGTTCGAGACCGAACGTGATCG-CACAATCCCAGTG 406
ACG10                   GATGGCGGCCATGATCCACGGGGACGTTCGAGACCGAACGTGATCG-CACAATCCCATGG 406
ACG3                    GATGGCGGCAATGATCCATGGGGATGTTCGAGACCGAACGTGAACG-CGCAATCCCATGG 406
ACG5                    GATGGCGGCCATGATCCAGCGGGACGTTCGAGACCGAACGTGATCG-CATAATCCCAATG 406
ACG2                    GATGGCGGCCACGATCTACGGGGACGTTCGAGACCAGACGTGATCG-CACAATCCCACGG 406
ACC2                    GATGGCGGCCACGATCTACGGGGACGTTCGAGACCGAACGTGATCG-CACAATCCCATGG 405
ACC6                    GATGGAGGCCATGATCCAGAGGGACGTTTGAGACCAGACGTGATCG-CATTGTCCCATGG 406
ACM-JQ910930.1          GATGGCGGCCACGATCCATAGGGATGTTCAAGAATAGACGTGCGCG-CGTTGTCCCACGG 406
ACM3                    GATGGCGGCCACGATCCATAGGGATGTTCAAGAATAGACGTGCGCG-CGTTGTCCCACGG 406
ACM5                    GATGGCGGCCACGATCCATAGGGATGTTCAAGAATAGACGTACGCG-CGTTGTCCCACGG 406
ACB-HF564640.1          GATGGCGGCCACGATCTATAGGGATGTTCAAGAATAGACGTGCACG-CGTTGTCCCACGG 406
ACB2                    GATGGCGGCCACGATCTATAGGGATGTTCAAGAATAGACGTGCACG-CGTTGTCCCACGG 406
ACB5                    GATGGCGGCCACGATCTATAGGGATGTTCAAGAATAGACGTGCACG-CGTTGTCCCACGG 406
ACM2                    GATGGCGGCAACGATCTATAGGGATGTTCAAGATTAGACGTGCACG-CGTTGTCCCACGG 406
ACB9                    GATGGCGGCCACGATCTATAGGGATGTTCAAGAATAGACGTGCACG-CGTTGTCCCACGG 406
ACM7                    GATGCCGGCAACAATCCATAGGGATGTTCAAGAATGGACGTGCCCG-CGTTGACCCACGG 406
ACM8                    GATGGCGGCCACGATCCATGGGGATGTTCAAGAATAGACGTGCGCG-CGTTGTCCCACGG 406
ACM10                   GATGGCCGCCACGATGCCTCGGGTTGTTCATGAAGAAACGTGCGCG-CGTTGACCCACGG 406
AcM4                    GATGGCGGCCACGATCCATAGGGACGTTCAAGAATAGACGTGCGCG-CGTTATCCCATGG 406
AcB4                    GATGGCGGCCACGATCCATAGGGATGTTCAAGAATAGACGTGCACG-CGTTGTCCCATGG 406
CU                      GACGACTGCCACGATCCACGGTGGT-TGGGCGAACATTCGCTCGAGGCCAAGACGTGCG- 415
CP                      GACGGCCGCCACGATCCACGGTGGA-TGAGTGATCACTCGCTCGAG-CCAGGTCGCGCG- 407
                        .* *   **.* .**  .    *      . **  . :**  . .* *   .:*  .

ACG-HF564641.1          GGAGGGTCGGACCCCGAATCCCACGGCGACCCCAG-TCAA------GCGG- 447
ACC1                    GGAGGGTCGGACCCCGAATCCCACGGCGACCCCAGGTCAG------GCGG- 450
ACC10                   GGAGGGTCGGACCCCGAATCCCACGGCGACCCCAGGTCAG------GCGG- 450
ACG1                    GGAGGGTCGGACCCCGAATCCCACGGCGACCCTAGGTCAG------GCGG- 450
ACC-HF564642.1          GGAGGGTCGGACCCCGAATCCCACGGCGACCCTAGGTCAG------GCGG- 449
ACG9                    GGAGGGTCGAACCCCGAATCCCACGGCGACCCTAGGTCAG------GCGG- 450
ACC3                    GGAGGGTCGGACCCCGAATCCCACGGCGACCCTAGGTCAG------GCGG- 450
ACC7                    GGAGGGTCGGACCCCGAATCCCTCCGCGACCCTAGGTCAG------GCGG- 450
ACC8                    GGAGGGTCGGACCCCGAATCCCACGGCGACACCAGGTCAG------GCGG- 450
ACG4                    GGAGGGTCGTACCGCGAATCCGACGGTGACCCCAGGTCAG------GCGG- 450
ACG10                   GGAGGGTCGGACCCCGAATCCCACGGCGACCCCAGGTCAG------GCGG- 450
ACG3                    GGAGGGCCGGACCCCGAATCCCACGGCAACCCCAGGTCAG------GCGG- 450
ACG5                    GGAGGGCGGGACCCCGAATCCCACGGCGACCCCAGGTCAG------GCGG- 450
ACG2                    GGAGGGTCGAACCCCGAATCCCACGGCGACCCTAGGTCAG------GCGG- 450
ACC2                    GGAGGGTCGGACCCCGAATCCCACGGCGACCCTAGGTCAG------GCGG- 449
ACC6                    GGAGGGCTGGACCCCGAATCCCACAGCAACCCCAGGTCAG------GCGG- 450
ACM-JQ910930.1          GGAGGGCTGAACCCCGAATCCCACAGCAACCCCAGGTCAG------GCGG- 450
ACM3                    GGAGGGCTGAACCCCGAATCCCACAGCAACCCCAGGTCAG------GCGG- 450
ACM5                    GGAGGGCCGAACCCCGAATCCCACAGCAACCCCAGGTCAG------GCGG- 450
ACB-HF564640.1          GGAGGGCCGAACCCCGAATCCCACAGCAACCCCAGGTCAG------GCGG- 450
ACB2                    GGAGGGCCGAACCCCGAATCCCACAGCAACCCCAGGTCAG------GCGG- 450
ACB5                    GGAGGGCCGAACCCCGAATCCCACAGCAACCCCAGGTCAG------GCGG- 450
ACM2                    GAAGGGCCGAACCCCGAATCCCACAGCAACCCCGGGTCAG------GCGG- 450
ACB9                    GGAGGGCCGAACCCCGAATCCCACAGCCACCCCAGGTCAG------GCGG- 450
ACM7                    GGAGGGTTGAACCCCGAATCCAACAGTAACCCCTGGTCAG------GCGG- 450
ACM8                    GGAAGGCCGAACCCCGAATCCCACAGCAACGCCAGGTCAG------GCGG- 450
ACM10                   GGAGGGCTGAACCCCGAATCCCACAGCAACCCCAGGTCAG------GCGG- 450
AcM4                    GGAGGGCTGAACCCCGAATCCCACAGCAACCCCAGGTCAG------GCGG- 450
AcB4                    GGAGGGCCGAACCCCGAATCCCACAGCAACCCCAGGTCAG------GCGG- 450
CU                      --CGGGTCGTGCCACG--------------GCAGGG----------GCC-- 438
CP                      --TGCGCCGTGCCACGGTGATGGCCCGGCAGCAAGGGCGCTCCGACGCCTT 456
                           . *  * .** **               *  *           **
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Table S3 Boxshade of Clustal W2 alignment with evidenced nucleotide differences

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ACG-HF564641.1     1 CAAGGAACTGCAACAGAGCGAG-AGCGCCTTACGCCGATTGCACCCGAAATGACTCTCAA
ACC1               1 ......................-..T........T.........................
ACC10              1 .................T....-.....................................
ACG1               1 ......................-.....T...............................
ACC-HF564642.1     1 ......................-.....................................
ACG9               1 ........C......A......-.A.....C.T...........................
ACC3               1 ........C......A......-CA.....C..............T..............
ACC7               1 ........A...........G.-.......A.....A.......................
ACC8               1 ......................-.....................................
ACG4               1 ......................-C....................................
ACG10              1 ......................-.....................................
ACG3               1 ......................-T....................................
ACG5               1 ...........G..........-...................................G.
ACG2               1 ..G...................-T......C.T.............A...C.........
ACC2               1 ......................-T......C.T.................C.......GG
ACC6               1 ....C.............G...-.......CTT.............A..TGC.....T..
ACM-JQ910930.1     1 ......................-T......C.T.............A...C.......GG
ACM3               1 ......................-T......C.T.............A...C.......GG
ACM5               1 ......................-T......C.T.............A...C.......GG
ACB-HF564640.1     1 ......................-T......C.T.............A...C.......GG
ACB2               1 ......................-T......C.T.............A...C.......GG
ACB5               1 ......................-T......C.T.............A...C.......GG
ACM2               1 ......................-T......C.T.............A...C.......GG
ACB9               1 ......................-TAG....C.T.............A...C.......GG
ACM7               1 A.C...................-T......C.T.............A...C.......GG
ACM8               1 ..................T...-G......C.T........G....A...C.......GG
ACM10              1 ....A...C......A......-C......C...............A....A......GG
AcM4               1 ......................-T......C.T.............A...C.......GG
AcB4               1 ......................-.......................A...C.......GG
CU                 1 .......AC.A....A...AG.GCA.T.G.---...AT.....T......C.......GG
CP                 1 ........A....AGA...A..GCA...A.G.T...AT.....T......C.......GG
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```
ACG-HF564641.1    60  CAACGGATATCTCAGCTCTCGCATCGATG-AGAACGTAGTGAAATGCAATGCTTGG
ACC1              60  .............................A..........................
ACC10             60  .............................A..........................
ACG1              60  .............................A..........................
ACC-HF564642.1    60  .............................A..........................
ACG9              60  .........................A...A.........C..........A.....
ACC3              60  T........................A...A.................G........
ACC7              60  .............................A.........G..........A.....
ACC8              60  .............................A..........................
ACG4              60  .............................A..........................
ACG10             60  .............................A..........................
ACG3              60  .........................A...A..........................
ACG5              60  .............................A...........G.....G........
ACG2              60  .............................A.....A...C.......G......A.
ACC2              60  .............................A.........C..............A.
ACC6              60  ..............CT...G.........A.......C.C................
ACM-JQ910930.1    60  .............G...............A.....A...C.......G........
ACM3              60  .............G...............A.....A...C.......G........
ACM5              60  .............G...............A.....A...C.......G........
ACB-HF564640.1    60  .............GA..............A.....A...C.......G......A.
ACB2              60  .............GA....T.G.......A.....A...C.......G......A.
ACB5              60  .............GA..............A.....A...C.......G......A.
ACM2              60  .............GA..............A.....A...C.......G......A.
ACB9              60  .............GA..............A.....A...C.......G........
ACM7              60  T.C..........G...............A.....A...C.......G........
ACM8              60  .............G...............A.....A...C......TG........
ACM10             60  .....T......T.......AT...T.AAA.....AG..C.....ATG..A.....
AcM4              60  ...A.........G...............A.....A...C................
AcB4              60  .............GA..............A.....A...C................
CU                58  .............G...............A.........C.......G..A.....
CP                61  .............G...............A.........C.......G..A.....
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ACG-HF564641.1   115 TGTGAATTGCAGAATCCTGTGAACTATCGAGTCTTTGAATGTAAGTTGCG-CCAAA
ACC1             116 ..................................................C.....
ACC10            116 ..................................................C.....
ACG1             116 ...........................T......................C.....
ACC-HF564642.1   116 ...........................T......................-.....
ACG9             116 .................C......C..G...................A..C..G..
ACC3             116 .................C......C..T.............C.....A..C.....
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```
ACC7            116 ...........A.....C.......................C........C.....
ACC8            116 ..................................................C.....
ACG4            116 ..................................................C.....
ACG10           116 ..................................................C.....
ACG3            116 ..................................................C.....
ACG5            116 ...........................T...................AT.C..C.T
ACG2            116 ...........................T...........C.C........C.....
ACC2            116 .................C......C..T.....C.....C.C......T.-.....
ACC6            116 ..........G........A.......T...........G.G..C...T.C.....
ACM-JQ910930.1  116 .................C......C..T.....C.....C.C......T.C..G..
ACM3            116 .................C......C..T.....C.....C.C......T.C..G..
ACM5            116 ...............T.C.G....C..T.....C.....C.C......T.C..G..
ACB-HF564640.1  116 .................C......C..T.....C.....C.C......T.C.....
ACB2            116 .................C......C..TT....C.....C.C......T.C.....
ACB5            116 .................C......C..T.....C.....C.C......T.C.....
ACM2            116 .................C......C..T.....C.....C.C......T.C.....
ACB9            116 .................C......C..T.....C.....C.C......T.C.....
ACM7            116 .................C......C..T.....C.....C.C......T.C..G..
ACM8            116 .....C..................C........C.....C.C......T.C..G..
ACM10           116 ......G..G.......C......C..G.............C.....A..C..G..
AcM4            116 ....................T...C..T.....C.....C.G......T.C..G..
AcB4            116 .................C......C..............C..........C.....
CU              114 .................C......C..............C.C........C..G..
CP              117 .................C......C..............C.C........C..G..

ACG-HF564641.1  170 GCTATTAGGCCGAGGGCACGCCTGCCTGGGCGTCACGCAACGTCGCCGCCACCCCA
ACC1            172 ........................................................
ACC10           172 ........................................................
ACG1            172 ........................................................
ACC-HF564642.1  171 ........................................................
ACG9            172 ..........T................A.........T...A..............
ACC3            172 .....................................T..................
ACC7            172 ..C......................T..............G...............
ACC8            172 ........................................................
ACG4            172 ..........................AT....A.......................
ACG10           172 ........................................................
ACG3            172 ...............................A...........T............
ACG5            172 ...................................G.................T..
ACG2            172 ..........................C....A...........T......G..T..
ACC2            171 ..C.......................C....A...........T......G..T..
ACC6            172 TTG.......G......C.......A.A...C.G.G......C......GC..T..
ACM-JQ910930.1  172 ..C.......................C....A.................T...T..
ACM3            172 ..C.......................C....A..................G..T..
ACM5            172 ..C..................G....C....A.................G...T..
ACB-HF564640.1  172 ..C.......................C....A...........T......G..T..
ACB2            172 ..C..................T....C....A...........T......G..T..
ACB5            172 ..C.......................C....A...........T......G..T..
ACM2            172 ..C.......................C....A...........T......G..T..
ACB9            172 ..C.......................C....A...........T......G..T..
ACM7            172 ..C........A...A.........T.....A.......T......GC.T...T..
ACM8            172 .TC.......................CTT........T.T.........GC..T..
ACM10           172 ..C.......T........C.................T........G..T...TG.
AcM4            172 ..C.......................C....C.....G..................
AcB4            172 ...............................A.....................T..
CU              170 ..CT................T.........T................AA.G...G.
CP              173 ..CT................T.........T........G.......A..G...G.

ACG-HF564641.1  226 CGCG--GGGCACGGCGAATGATGGCCTCCCGAGGCCTCTATCCCCCAGCTGGCCGA
ACC1            228 ....--..................................................
ACC10           228 ....--..................................................
ACG1            228 ....--..................................................
ACC-HF564642.1  227 ....--..................................................
ACG9            228 ....--........T........................G.............T..
ACC3            228 ....--..................................................
ACC7            228 ....--..................................................
ACC8            228 ....--..................................................
ACG4            228 ....--.A......................AGA.G..........T.A........
ACG10           228 ....--........................A................A........
ACG3            228 ....--....C...T.......................C.................
ACG5            228 ....--....G.C..A..............CGA.G.................A...
ACG2            228 ....--........................................G.........
ACC2            227 ....--A...G....................G......C......T....A.....
ACC6            228 ....--....G.C..AG........G........G...........G...A..G..
ACM-JQ910930.1  228 ....--....G...TAG............TAG..G...C......TG...A..T..
ACM3            228 ....--....G...TAG............TAG..G...C......TG...A..T..
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ACM5            228 ....--....G...TAG............TAG..G...C......TG...A..T..
ACB-HF564640.1  228 ....--....G...TAG.............AG..G...C......TG...A..T..
ACB2            228 ....--....G...TAG.............AG..G...C......TG...A.GT..
ACB5            228 ....--....G...TAG.............AG..G...C......TG...A..T..
ACM2            228 ....--....G..ATAG......A......AG..G...G......TG...A..T..
ACB9            228 ....--....G...TAG.............AG..G...C......TG...A..T..
ACM7            228 ....--....G...TAG.A......T..TTAG......C......TG...ACTT..
ACM8            228 A...--....G...TAG...........TTTG..G...C....T..G...A.....
ACM10           228 ....--....G...TTG............TAG..G...C......TG...A..T..
AcM4            228 ....--....GG..T.G.............AG..G...C.......G......T..
AcB4            228 ....--....G...TAG.............AG..G...C......AG......T..
CU              226 TC.TCT....GT....G..............G.AG.CTCGC.T...G.........
CP              229 TCATGC....TT....G..............G.AG.GTCGC.T...G.........
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ACG-HF564641.1  280 AATAACTGCCTAGCGCGATGGCGGCAATGATCCACGGGGACGTTCGAGACCGAACG
ACC1            282 .........................C..............................
ACC10           282 .........................C..............................
ACG1            282 .........................C..............................
ACC-HF564642.1  281 .........................C..............................
ACG9            282 ...G..G...........C..T...C.C........A.....C.T...........
ACC3            282 .........................C..............................
ACC7            282 ................A........C...........A..A...............
ACC8            282 .........................C..............................
ACG4            282 .........................C..........T...................
ACG10           282 .........................C..............................
ACG3            282 ...........C......................T.....T...............
ACG5            282 ...C.....................C........GC....................
ACG2            282 ...........C.............C.C....T..................AG...
ACC2            281 ...G..G..................C.C....T.......................
ACC6            282 ...G...C.......T.....A...C........GA........T......AG...
ACM-JQ910930.1  282 ...G.TG....C.............C.C......TA....T....A...ATAG...
ACM3            282 ...G.TG....TA............C.C......TA....T....A...ATAG...
ACM5            282 ...G.TG..................C.C......TA....T....A...ATAG...
ACB-HF564640.1  282 ...G.TG....C.............C.C....T.TA....T....A...ATAG...
ACB2            282 ...G.TG....C.............C.C....T.TA....T....A...ATAG...
ACB5            282 ...G.TG....CA............C.C....T.TA....T....A...ATAG...
ACM2            282 ...G.TG....C...............C....T.TA....T....A...TTAG...
ACB9            282 ...G.TG....C.............C.C....T.TA....T....A...ATAG...
ACM7            282 .T.G.TGC...CC.C.....C......CA.....TA....T....A...AT.G...
ACM8            282 ...G.TG......T...........C.C......T.....T....A...ATAG...
ACM10           282 ...GTTG....C..........C..C.C...G.CTC...TT....AT..AGA....
AcM4            282 ...G.TG....C.............C.C......TA.........A...ATAG...
AcB4            282 .....TG..................C.C......TA....T....A...ATAG...
CU              282 ..A.GGG...CGA..T..C.A.T..C.C.........T.GT-.GG.C..A.ATT..
CP              285 ....GGG...C.A..T..C...C..C.C.........T.GA-.GA.T..T.ACT..
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ACG-HF564641.1  336 TGATCG-CACAATCCCATGGGGAGGGTCGGACCCCGAATCCCACGGCGACCCCAG-
ACC1            338 ......-................................................G
ACC10           338 ......-................................................G
ACG1            338 ......-.............................................T..G
ACC-HF564642.1  337 ......-.............................................T..G
ACG9            338 ......-......................A......................T..G
ACC3            338 ......-.............................................T..G
ACC7            338 ......-...................................T.C.......T..G
ACC8            338 ......-...........................................A....G
ACG4            338 ......-..........GT..........T...G.......G....T........G
ACG10           338 ......-................................................G
ACG3            338 ...A..-.G.................C....................A.......G
ACG5            338 ......-..T.......AT.......CG...........................G
ACG2            338 ......-..........C...........A......................T..G
ACC2            337 ......-.............................................T..G
ACC6            338 ......-..TTG..............CT................A..A.......G
ACM-JQ910930.1  338 ..CG..-.GTTG.....C........CT.A..............A..A.......G
ACM3            338 ..CG..-.GTTG.....C........CT.A..............A..A.......G
ACM5            338 .ACG..-.GTTG.....C........C..A..............A..A.......G
ACB-HF564640.1  338 ..CA..-.GTTG.....C........C..A..............A..A.......G
ACB2            338 ..CA..-.GTTG.....C........C..A..............A..A.......G
ACB5            338 ..CA..-.GTTG.....C........C..A..............A..A.......G
ACM2            338 ..CA..-.GTTG.....C...A....C..A..............A..A.....G.G
ACB9            338 ..CA..-.GTTG.....C........C..A..............A..C.......G
ACM7            338 ..CC..-.GTTGA....C.........T.A...........A..A.TA.....T.G
ACM8            338 ..CG..-.GTTG.....C.....A..C..A..............A..A..G....G
ACM10           338 ..CG..-.GTTGA....C........CT.A..............A..A.......G
AcM4            338 ..CG..-.GTT...............CT.A..............A..A.......G
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AcB4             338 ..CA..-.GTTG..............C..A..............A..A.......G
CU               337 CTCGA.G.CA.GA.GTGC.---C......TG..A..--------------G.AG.G
CP               340 CTCGA.-.CAGG..G.GC.---T.C.C..TG..A..GTGATGG.CCG.CAG.A..G
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ACG-HF564641.1  390 TCAA------GCGG- 447
ACC1            393 ...G------....- 450
ACC10           393 ...G------....- 450
ACG1            393 ...G------....- 450
ACC-HF564642.1  392 ...G------....- 449
ACG9            393 ...G------....- 450
ACC3            393 ...G------....- 450
ACC7            393 ...G------....- 450
ACC8            393 ...G------....- 450
ACG4            393 ...G------....- 450
ACG10           393 ...G------....- 450
ACG3            393 ...G------....- 450
ACG5            393 ...G------....- 450
ACG2            393 ...G------....- 450
ACC2            392 ...G------....- 449
ACC6            393 ...G------....- 450
ACM-JQ910930.1  393 ...G------....- 450
ACM3            393 ...G------....- 450
ACM5            393 ...G------....- 450
ACB-HF564640.1  393 ...G------....- 450
ACB2            393 ...G------....- 450
ACB5            393 ...G------....- 450
ACM2            393 ...G------....- 450
ACB9            393 ...G------....- 450
ACM7            393 ...G------....- 450
ACM8            393 ...G------....- 450
ACM10           393 ...G------....- 450
AcM4            393 ...G------....- 450
AcB4            393 ...G------....- 450
CU              376 ----------..C-- 438
CP              392 G.GCTCCGAC..CTT 456
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Table S4 Estimates of Genetic Distance over Sequence Pairs between Groups

The number of base substitutions per site from averaging over all sequence pairs between groups are shown. Analyses were conducted using the Maximum Composite Likelihood model in MEGA6. The analysis involved 29 nucleotide sequences.

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M, ,
G,0.191, ,
C,0.203,0.08, ,
B,0.07,0.163,0.174, ,
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