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Phylogenetic placement of owl monkey, genus *Aotus*, using *Alu* retrotransposons

by

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Undergraduate honors thesis under the direction of

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Abstract

Looking for the presence of shared *Alu* insertions in the genomes of primates within the New World monkey clade Cebidae presents itself as a promising method to resolve the phylogenetic tree of Cebidae. *Aotus* is one of the genera within this clade having an unresolved phylogenetic placement due to the rapid divergence of owl monkeys (*Aotus*) and the other members of Cebidae, mainly marmoset, squirrel monkey, and capuchins. *Alu* elements are transposable elements, or stretches of DNA that get copied and pasted throughout the genome via an RNA intermediate. The more time that passes throughout evolutionary time scales, the more time the genomes have to accumulate these insertions, which leads us to the idea that the ancestral state would lack a particular *Alu* element compared to other genomes that inherited the insertion more recently. Using the findings from genomic analysis [including sequence analysis and Polymerase Chain Reaction] of the genomes, we can try to determine how the genera within Cebidae were derived. This study examined *Alu* insertions shared by three Cebidae genomes and absent in a fourth to resolve the branching order of *Aotus* within Cebidae. The results indicate that *Aotus* and marmoset diverged at nearly the same time and that both are basal to squirrel monkey and capuchins.

Introduction

Mobile elements serve a role in human genomic diversity and make up almost half of the human genome by mass (Batzer & Deininger, 2002). They serve an equally large presence in non-human primates, and this is due to the spread of repeats throughout these genomes (Konkel,

Walker, & Batzer, 2010). Their presence can alter gene expression if inserted into a protein coding region via recombination processes such as deletions, duplications, or inversions (Cordaux & Batzer, 2009). This genomic instability created by their large increase in copy number is due to their selfish and almost parasitic nature (Cordaux & Batzer, 2009). Among these mobile elements are LINEs (long interspersed elements) and SINEs (short interspersed elements). Within the category of SINEs, which are less than 500 DNA base pairs (bp) long, fall the *Alu* elements, DNA stretches of approximately 300 bp that are amplified through retrotransposition via RNA polymerase-mediated reverse transcription (Batzer & Deininger, 2002). Mobile elements can follow a cut and paste or a copy and paste procedure, but the most common type in mammals is copy and paste via an RNA intermediate (Konkel et al., 2010). They are further classified as either autonomous or non-autonomous based on whether or not they have the necessary equipment built in (Konkel et al., 2010). *Alu* elements themselves lack their own open reading frames and are therefore non-autonomous and rely on the machinery of LINEs, meaning they use a common pathway for propagation (Batzer & Deininger, 2002). This characteristic of *Alu* elements has earned them the name of the “parasite’s parasite” since they rely on the machinery of another “parasite”, or mobile element, to fulfill their selfish need to spread in the genome (Cordaux & Batzer, 2009). Gene function is interrupted due to mutations that are potentially lethal as a result of the insertion of *Alu* elements into coding regions of DNA, which accounts for the fact that they are rarely found within coding regions (Callinan & Batzer, 2006). The retrotransposons L1 (LINE) and *Alu* (SINE) have shaped primate genome structure and evolution, and due to the presence of flanking target site duplications (TSDs), we can see where they have been copied and pasted into the genome (Konkel et al., 2010). These TSDs are located on both the 5' and 3' ends and are a product of movement via TPRT (target site-primed

reverse transcription) (Luan, Korman, Jakubczak, & Eickbush, 1993; J. M. Storer, Walker, Jordan, & Batzer, 2020). Shared insertions of retrotransposons serve as strong indicators of common ancestry between species or individuals because they are considered homoplasy-free and identical by descent (Konkel et al., 2010). This is why the use of *Alu* elements can help resolve the phylogenetic history among primates. While both *Alu* and L1 elements are common in primate genomes, it is more practical to study *Alu* elements when genotyping because of their relatively small size; L1 insertion size can reach 6 kb compared to a normal *Alu* insertion length of 300 bp (Konkel et al., 2010). Another supporting factor in using *Alu* elements in the study of primate evolution is that the origin and amplification of *Alu* elements and the radiation of primates occurred around the same time about 65 million years ago (mya) (Callinan & Batzer, 2006).

The platyrhines, or New World monkeys (NWM), consists of three recognized families: Cebidae, Atelidae, and Pitheciidae. The Cebidae family contains four subfamilies: Callithrichinae (marmosets and tamarins), Cebinae (*Cebus* and *Sapajus* capuchins), Saimirinae (squirrel monkeys), and Aotinae (owl monkeys). Within the Cebidae family lies controversy over the placement of owl monkey, genus *Aotus*, because of ambiguity due to rapid speciation (J. M. Storer et al., 2020). Approaches to studying the phylogeny of platyrhines using the fossil record have been useful in uncovering and describing the diversity in physical characteristics but have not always been easily accessible and as efficient as molecular analysis at reconstructing phylogeny (Aristide, Rosenberger, Tejedor, & Perez, 2015). Morphology has not been a reliable basis of settling the phylogeny of New World monkeys because the conditions of South America and the Amazon region where these primates have lived has not been conducive to fossil preservation (J. M. Storer et al., 2020). However, studies using the fossil record have come to

similar conclusions in finding a close relationship between *Aotus* and Callitrichinae within the Cebidae family (Aristide et al., 2015). Thus, the genome and its molecular markers have been key in reconstructing the NWM phylogeny (Osterholz, Walter, & Roos, 2009; Ray et al., 2005; J. M. Storer et al., 2020). It is thanks to these studies that there is an agreement on the three families of platyrhines as listed above (J. M. Storer et al., 2020). *Alu* lineages can allow us to understand the phylogeny of Cebidae because *Alu* elements accumulate in an identical by descent manner. Around the divergence of New World and Old World monkeys, or platyrhines and catarrhines, there were three primary *Alu* lineages: *AluJ*, *AluS*, and an *AluY* progenitor that became *AluY* only in catarrhines after the split. In platyrhines, *AluSp* and *AluSc* were active and led to the *AluT* lineage found only on platyrhines (Ray & Batzer, 2005). *AluT* represents a gene conversion event (between *AluSp* and *AluSc*) and a sudden incorporation of unique mutations, which is special because it did not require gradual accumulation and is relatively recent (Ray & Batzer, 2005). The platyrhine *AluT* lineage evolved into three subfamilies which are strongly supported and accepted: *AluTa7*, *AluTa10*, and *AluTa15* (Ray & Batzer, 2005). *Ta7* and *Ta10* are found in all three platyrhine families, while *Ta15* is unique to Cebid taxa (Ray & Batzer, 2005). Upon insertion, *Alu* copies remain in the genomes of descendants as what are considered to be perfect homologies (Schneider & Sampaio, 2015). This is another supporting factor in the identical-by-descent nature of *Alu* elements that makes them a good tool in genomic studies of phylogeny: if an *Alu* insertion is shared by two taxa, they must share a common ancestor that passed it on to them (Schneider & Sampaio, 2015).

The accumulation of these insertions is very helpful in phylogenetic and evolutionary studies due to the insertions being identical-by-descent and homoplasy-free (Cordaux & Batzer, 2009), and a notable study on the human-chimpanzee-gorilla phylogeny used *Alu* elements to

resolve this trichotomy (Konkel et al., 2010; Salem et al., 2003). Because homoplasy refers to similarity not due to a common ancestor, being homoplasy-free means that their similarity is due to having a common ancestor. For each locus in the genome, there is the possibility of presence or absence of the insertion, and the absence indicates the ancestral state since these are derived insertions (Cordaux & Batzer, 2009). The presence of one of these insertions would thus indicate the more recently diverged group. Whole genome sequence (WGS) based genomic analyses allow for the identification of the specific location on the chromosome of the *Alu* insertion (J. M. Storer et al., 2020). Using the characteristics that we know about *Alu* elements such as their 300 bp length, GGCCGGG start sequence, and poly A tail, we can study the position of the highly contested *Aotus* genus within the Cebidae family (J. M. Storer et al., 2020). In this study, we designed primers localized to the DNA regions that appear to contain shared *Alu* insertions after looking at multi-sequence alignments and performed PCR amplification on these DNA samples to confirm or deny presence of these identical-by-descent elements. A large number of *Alu* insertions shared by marmoset, squirrel monkey, and capuchin genomes to the exclusion of owl monkey would indicate that *Aotus* is basal within Cebidae, whereas evidence to the contrary would imply that *Aotus* is not basal or still remains unsolved.

Materials and Methods

INITIAL IDENTIFICATION OF ALU ELEMENT PRESENCE

Four-way sequencing alignments for Cebidae genomes were constructed by Jessica Storer during her doctoral dissertation in Dr. Batzer's laboratory (J. Storer, 2019; Jessica M. Storer et al., 2019). The four genomes obtained from NCBI were (M): common marmoset;

[caljac3], (C): capuchin monkey; [Cebus_imitator-1.0], (S): squirrel monkey; [sajBol1], and (O): owl monkey; [Anan_2.0]. They were analyzed for their *Alu* content using RepeatMasker (RepeatMasker-Open-4.0) (Smit, Hubley, & Green, 2015). Full-length elements, with 600 bp of 5' and 3' flanking sequence, for each genome were then aligned against the other three genomes. These methods are described in detail in Storer et al. 2020. The datasets provided for this study consisted of four-way sequence alignments in which an *Alu* insertion appeared to be shared in three genomes and absent from the fourth. These four categories were 1). COS, shared by capuchin, owl monkey, and squirrel monkey to the exclusion of marmoset; 2). CMS, shared by capuchin, marmoset, and squirrel monkey to the exclusion of owl monkey; 3). MOS (absent in capuchin); or 4). CMO (absent in squirrel monkey). Following visual inspection of alignments, oligonucleotides for PCR were attempted for those that passed inspection. A summary is shown in Table 1.

DNA PANEL

The DNA panel used contained samples from 19 individuals, including both New World monkeys and Old World monkeys. The species used included the following: human, *Homo sapiens* (1 sample); common chimpanzee, *Pan troglodytes* (1 sample); and African green monkey, *Chlorocebus aethiops* (1 sample). New World monkeys representing the Atilidae family included Woolly monkey, *Lagothrix lagotricha* (1 sample); spider monkeys, *Ateles belzebuth* (1 sample) and *Ateles geoffroyi* (1 sample); and howler monkey, *Alouatta sara* (1 sample). The Cebidae family had DNA samples for common marmoset, *Callithrix jacchus* (1 sample); Pygmy marmoset, *Callithrix pygmea* (1 sample); Goeldi's marmoset, *Callimico goeldii* (1 sample); tamarins, *Saguinus labiatus* (1 sample) and *Saguinus fuscicollis nigrifrons* (1 sample).

sample); *Sapajus apella* capuchins (3 samples); squirrel monkey, *Saimiri s. sciureus* (1 sample); and owl monkey, *Aotus trivirgatus* (1 sample). The Pitheciidae family had representative DNA samples from saki, *Pithecia p. pithecia* (1 sample), and titi, *Callicebus d. donacophilus* (1 sample). Among this DNA panel, we have humans, Old World monkeys, and New World monkeys from Atelidae, Cebidae, and Pitheciidae. The samples were obtained from various research centers. A full chart of these DNA samples is found in Appendix 1.

OLIGONUCLEOTIDE PRIMER DESIGN

The four Cebidae genomes were analyzed one-by-one at many genomic loci to search for the presence of shared *Alu* insertions. Candidates that appeared to contain these 300 bp insertions at the same insertion site were used to make oligonucleotide primers for PCR from the homologous flanking sequence region. The primers were accompanied by predicted PCR product size, which was used for comparison when interpreting the results of PCR.

Primers obtained from Sigma-Aldrich in pelleted oligonucleotide form were reconstituted to a final stock concentration of 100 µM. This was done by adding the manufacturer-determined amount of 1x TLE (TLE: 10mM Tris / 0.1 mM EDTA). To obtain a working concentration of 2 µm for each primer pair, 10 µL of forward primer and 10 µL of reverse primer were added to 480 µL of 1x TLE. Primers and predicted amplicon fragment sizes are available in the Appendix 2.

PCR

Polymerase Chain Reaction (PCR) was used to determine whether the *Alu* elements were present or absent inside each stretch of the genome of various species according to the different DNA primers used. (Reactions were carried out in groups of 24, one for each species and control.) The PCR mix used contained concentrations of 25-50 ng of DNA, 200 nM of forward and reverse primer, 0.2 mM deoxyribonucleotide triphosphates, 1.5 mM MgCl₂, 10X PCR buffer, and 1 unit of *Taq* DNA polymerase. The total volume of each well used for amplification was 25 µL, and the following conditions were used: 95°C for 1 minute, 32 cycles of 30 seconds of denaturation, annealing, and extension with denaturation at 95°C, annealing at 57-60°C, and extension at 72°C. This was then finished with an extension step at 72°C for 2 minutes.

After amplification, a 2% agarose gel with 0.2 µg/mL ethidium bromide was used for gel electrophoresis of the samples. An electric current of 170 V was applied to the DNA in the gel for 60 minutes and then observed using UV fluorescence made possible by the addition of ethidium bromide. The negative charge of DNA causes it to move down the gel through the agarose matrix towards the positive electrode, with smaller segments of DNA moving faster. The distance on the gel traveled by the DNA allows us to determine their relative sizes using a 100 bp DNA ladder to determine if there is an *Alu* insertion, or a fragment that is 300 base pairs shorter.

ALU INSERTION POLYMORPHISM

To interpret the results of PCR, we compared the sizes of the DNA amplified to assign each individual in its respective DNA sample a code of (1,1) for homozygous present for the *Alu* locus, (0,0) for homozygous absent for the *Alu* locus, (1,0) for heterozygous for the *Alu* locus, or

(-9,-9) in cases of no amplification or bands detected on the gel. These designations were recorded onto an Excel file, and the different species were compared. This information may be found in Appendix 3.

Results

This study attempted to resolve the phylogenetic placement of owl monkeys, genus *Aotus*, within the Cebidae family of New World monkeys using hundreds of potentially shared *Alu* insertions. The results of the four-way alignments are summarized in Table 1. A total of 875 *Alu* insertions were examined by 4-way alignment among the 4 possible categories as described in the methods. Only about 10-20% of them actually appeared to be shared by three species and absent from the 4th genome after visual inspection. Many of the candidates had N's in the sequence, had truncated sequences for one or more of the genomes, or had portions of the *Alu* in all four species of the alignments. These cases were excluded from further experiments. Examples are shown in Figure 1. PCR analyses were conducted on all remaining candidates that PCR primers could be designed for, and at least half from each category (n=98 total) with emphasis on CMS (absent from owl monkey, suggesting that *Aotus* is basal within Cebidae) or COS (absent from marmoset, suggesting that Callitrichinae—marmoset and tamarin—are basal within Cebidae), as these two categories were most likely to be true based on the number of alignment-producing candidates.

The COS category has the highest numbers with 63 *Alu* elements passing alignment inspection out of 286 candidates and 16 that were confirmed by PCR out of 40 tested by gel electrophoresis. Figure 2A. These results indicate that marmoset is most basal in Cebidae out of

the four genomes in this study. However, the CMS category placing *Aotus* basal to the other three genomes is almost as likely with 14 confirmed by PCR out of 32 (Figure 2B), with a validation rate of 44% compared to 40% for the COS group. The other two categories, MOS and CMO, meaning that capuchin or squirrel monkey is basal, respectively, have far less support and are considered very unlikely based on phylogenetic studies. However, the fact that all four categories have PCR confirmed shared *Alu* insertions (Figure 2) suggests that the radiation of these genera was very rapid, causing extensive incomplete lineage sorting of *Alu* elements that had not yet reached fixation. PCR results that did not confirm the predicted relationship most often showed the *Alu* present in all NWM samples or had poor amplification.

Table 1. Summary of four-way alignments visual inspection and PCR results for each category

Shared category	CMO	CMS	COS	MOS	All
Unique <i>Alu</i> alignments	192	238	286	159	875
Post-align NP	3	5	2	4	14
Post-align other*	169	181	221	132	703
Post-align shared	20	52	63	23	158
Analyzed by PCR	10	32	40	16	98
PCR confirmed	4	14	16	3	37
% confirmed by PCR	4/10 (40%)	14/32 (44%)	16/40 (40%)	3/16 (19%)	37/98 (38%)

NP: Near-parallel insertion; * N's in sequence, truncations or portion of *Alu* in all 4

Figure 1A: CMS-20

610 620 630 640 650 660 670 680 690 700
|.....|.....|.....|.....|.....|.....|.....|.....|.....|
 20_C **TGCTAAAGATTTAAAAGCAAGactagttggcgcacatggctccatcgatgtttgggggttgatggggggatggatcaacgaa**
 20_M **TGCTAAAGATTTAAAAGTGAGaatatgcgggttcgcgttgcacatcgatgtttgggggttgatggggggatggatcaacgaa**
 20_S **TGCTAAAGATTTAAAGAGTGAGAataatgcgggttcgcgttgcacatcgatgtttgggggttgatggggggatggatcaacgaa**
 20_O **TGCTAAAGATTT-**
|.....|.....|.....|.....|.....|.....|.....|.....|
 710 720 730 740 750 760 770 780 790 800
|.....|.....|.....|.....|.....|.....|.....|.....|
 20_C **tgcggaccatcctggtaacatggtgaaacccccgttcctactgaaaaatacaaaaaattatggatggcatggttggccacac**
 20_M **tgcggaccatcctggtaacatggtgaaacccccgttcctactgaaaaatacaaaaaattatggatggcatggttggccacac**
 20_S **tgcggaccatcctgtatcaacatggtgaaacccccgttcctactgaaaaatacaaaaaattatggatggcatggttggccacac**
 20_O **-**
|.....|.....|.....|.....|.....|.....|.....|.....|
 810 820 830 840 850 860 870 880 890 900
|.....|.....|.....|.....|.....|.....|.....|.....|
 20_C **gaactgtgcaggagaattgttgcacccaggaggcaggggttgcgttgcgttgcacccatgcgcattgcactccagccatggcaaca**
 20_M **gaactgtgcaggagaattgttgcacccaggaggcaggggttgcgttgcgttgcacccatgcgcattgcactccagccatggcaaca**
 20_S **gaactgtgcaggagaattgttgcgcattgcactccatgcgcattgcactccatgcgcattgcactccatgcgcattgcactccat**
 20_O **-**
|.....|.....|.....|.....|.....|.....|.....|.....|
 910 920 930 940 950 960 970 980 990 1000
|.....|.....|.....|.....|.....|.....|.....|.....|
 20_C **tctc-aaaaaaaaaaaaaaaatgtgagaataaGAACCTAATGCCTTCAGGGAACTCATTAAGTCAGTGAGGAAGGCAGATGTGAACAAATAATTGAA**
 20_M **tgtttt---agaaaaaaaaaaaatgtgagaataaGAACATTAATGCCTTCAGGGAACTCATTAAGTCAGTGAGGAAGACAGATGTGAACAAATAATTGAA**
 20_S **tgtctcaaaaaaaaaaaaaaaatgtgagaataaGAACCTAATGCCTTCAGGGAACTCATTAAGTCAGTGAGGAAGACAGATGTGAACAAATAATTGAA**
 20_O **AAAAGT GAGAATAAAGAACCTAATGCCTTCAGGGAACTCATTAAGTCAGTGAGGAAGACAGATGTGAACAAATAATTGAA**

Figure 1B: CMS-11

Figure 1. Genome alignments showing shared *Alu* insertions in capuchin, marmoset, and squirrel monkey to the exclusion of owl monkey at two different candidate loci. Figure 1A shows locus CMS-20, and Figure 1B shows locus CMS-11. In Figure 1A, the beginning of the *Alu* insertion as identified by the start sequence “ggccggg” can be found at the 625 bp mark with a sequence of “gccggg”. This was not uncommon to have start sequences that differed slightly from what was expected. The start region when a gap in the sequence of the excluded genome is seen (613 bp

mark); in this case it was that of the owl monkey, which shows “aaaagtgagaata” before the *Alu*. The termination of this insertion where we see the poly-A tail is around the 910 bp mark for a total *Alu* insertion link of close to 300 bp as expected. At the 918 bp mark is shown the same TSD, “aaaagtgagaata”, that was used to determine a shared insertion. The fourth row, which is the owl monkey, lacks the *Alu* insertion, suggesting that it is basal and would have diverged before the introduction of this identical by descent insertion. In Figure 1B, the *Alu* insertion appears to begin around the 627 bp mark with a start sequence of “agccagg”. This is another example of the start sequence differing slightly from the expected start sequence of “ggccggg”. One thing that can be observed in this alignment is that the owl monkey genome is missing a section of only about 130 bp in length, instead of the expected 300 bp. In fact, what is observed is that the owl monkey genome contains some of the *Alu* insertion. As mentioned, the beginning of this insertion begins at the 627 bp mark, but the owl monkey genome has bases leading all the way to the 651 bp mark. In addition, the poly-A tail marking the end of the *Alu* insertion is found at about the 920 bp mark, but the owl monkey genome has bases appearing again at the 784 bp mark. The TSD “aaagagcagccat” appears at both ends of the *Alu* in all 4 sequences. Therefore, Figure 1B represents an alignment that would be interpreted as having parts of the *Alu* element in all 4 of the genomes being compared, and it, thus, does not present itself as a strong candidate for primer selection since the owl monkey genome is not a true exclusion.

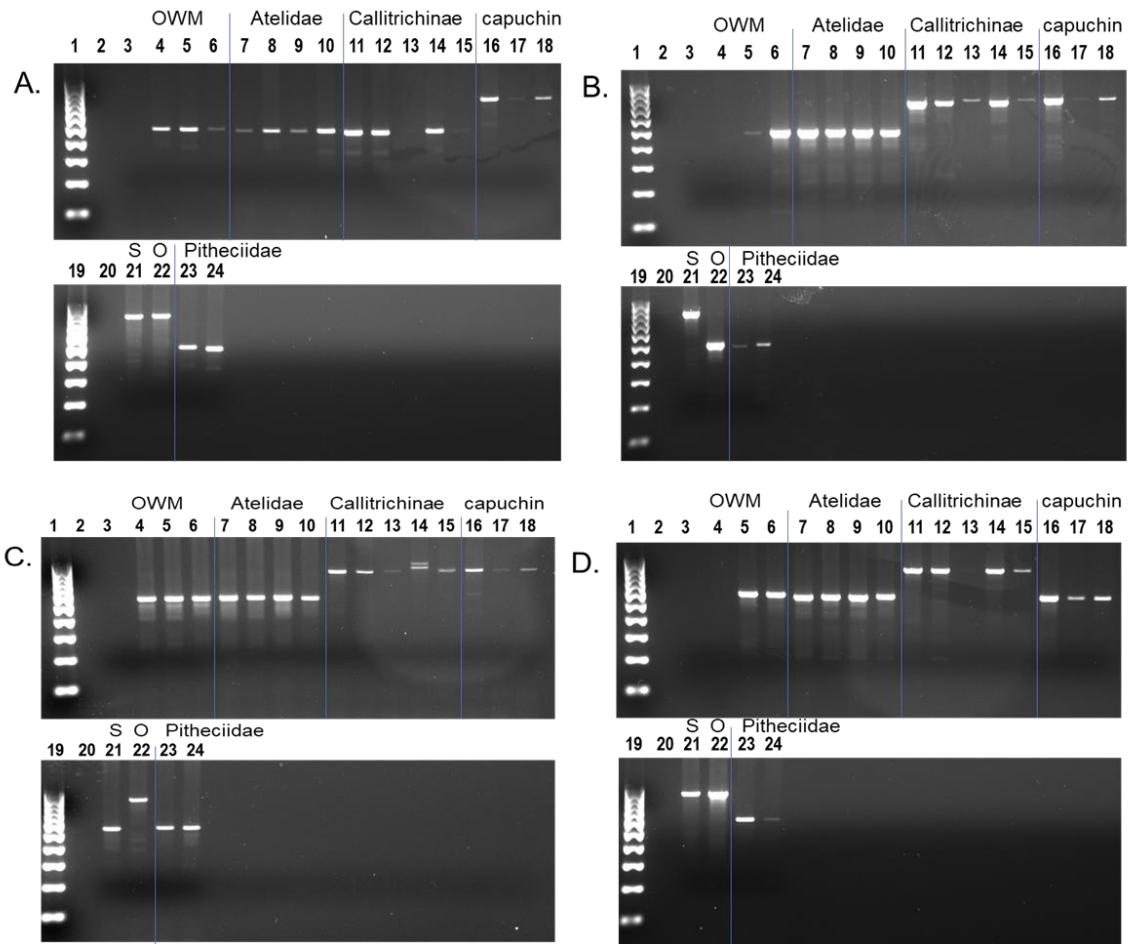


Figure 2. PCR analyses of *Alu* elements shared in three of four Cebidae genera and also absent in Atelidae and Pitheciidae. Lanes: 1- 100 bp DNA ladder, 2- blank, 3- TLE (negative control), 4- Human (HeLa), 5- *Pan troglodytes* (common chimpanzee), 6- *Chlorocebus aethiops* (African green monkey), 7- *Lagothrix lagotricha* (woolly monkey), 8- *Ateles belzebuth* (white bellied spider monkey), 9- *Ateles geoffroyi* (black-handed spider monkey), 10- *Alouatta sara* (Bolivian red howler monkey), 11- *Callithrix jacchus* (common marmoset), 12- *Callithrix pygmea* (Pygmy marmoset), 13- *Callimico goeldii* (Goeldi's marmoset), 14- *Saguinus labiatus* (red-chested mustached tamarin), 15- *Saguinus fuscicollis nigrifrons* (Geoffroys saddle-back tamarin), 16-18 *Sapajus apella* (tufted capuchin), 19- 100 bp DNA ladder, 20- blank, 21- *Saimiri s. sciureus* (common squirrel monkey), 22- *Aotus trivirgatus* (Three striped owl monkey), 23- *Pithecia p. pithecia* (Northern white-faced saki), 24- *Callicebus d. donacophilus* (Bolivian gray titi monkey). A lighter molecular weight as identified by the presence of the band lower on the gel signifies a smaller fragment, and, thus, a sample lacking the 300 bp *Alu* insertion. A) COS #51, *Alu* is present in capuchin, owl monkey and squirrel monkey (~790 bp DNA fragment lanes 16-18, 21-22) and absent in marmosets and tamarins (~465 bp DNA fragment lanes 11-15). This *AluSc* supports callitrichines (marmosets and tamarins) as basal within Cebidae. B) CMS #64, *Alu* is present in capuchin, marmosets, tamarins and squirrel monkey (~800 bp DNA fragment lanes 11-18, 21) and absent in owl monkey (~496 bp DNA fragment lane 22). This *AluSc* supports owl monkey (*Aotus*) as basal within Cebidae. C) CMO #29, *Alu* is present in capuchin, marmosets,

tamarins and owl monkey (~860 bp DNA fragment lanes 11-18, 22) and absent in squirrel monkey (~525 bp DNA fragment lane 21). This *Alu*Ta7 supports squirrel monkey as basal within Cebidae. D) MOS #5, *Alu* is present in marmosets, tamarins, owl monkey and squirrel monkey (~880 bp DNA fragment lanes 11-15, 21-22) and absent in capuchins (~564 bp DNA fragment lanes 16-18). This *Alu*Ta7 supports capuchin as basal within Cebidae.

Subfamilies *Alu*Sc and *Alu*Ta7 were both active simultaneously with the divergence of platyrhines and catarrhines (Ray & Batzer, 2005) about 20 mya. The Ta-lineage is unique to platyrhines. These data are consistent with incomplete lineage sorting (ILS) of *Alu* insertions that occurred after the Cebidae divergence from Atelidae and Pitheciidae but immediately prior to the rapid speciation of Cebidae taxa. These *Alu* elements remained unfixed within Cebidae and became randomly assorted for presence or absence in subsequent emerging species.

Discussion

Previous studies have attempted to resolve the placement of *Aotus* within Cebidae using a few *Alu* insertions (Osterholz et al., 2009; Ray et al., 2005) or other methods (Jameson Kiesling, Yi, Xu, Gianluca Sperone, & Wildman, 2015; Valencia, Martins, Ortiz, & Di Fiore, 2018), and none have resulted in complete agreement. That is why this study started by mining all possible *Alu* insertions from the available genome assemblies and aligning them to the other three genomes to get as many candidates as possible. Still, both COS and CMS seem equally likely, thus indicating that the radiation of *Aotus* and the Callitrichinae (leading to marmoset and tamarins) occurred at nearly the same time. Rapid radiation about 20 mya resulted in extensive incomplete lineage sorting of *Alu* elements that were not fixed in the various populations at the time of speciation and became randomly assorted for presence or absence in these Cebidae lineages. Shared *Alu* insertions in this study were often identified as *Alu*Sc or *Alu*Ta7 subfamilies

which were mobilizing at the time of radiation. These insertions are roughly 20 million years old, and while some sequence decay was observed in the alignments, the *Alu* start and stop positions and TSDs could usually be identified.

While the relationship among the New World monkey families Cebidae, Atelidae, and Pitheciidae are supported as monophyletic, the relationship among the genera of Cebidae remains controversial because when studying the genomes, we sometimes get contradictory results. In one study, SINE insertions were used to solve this relationship, but the results found a sister grouping of *Aotus* and *Saimiri* and of *Saimiri* and *Cebus*, likely the result of the incomplete lineage sorting that has made the reconstruction of this phylogeny so difficult (Osterholz et al., 2009). Other studies have highlighted the problematic position of *Aotus* within Cebidae and have explored different techniques such as Restriction-site Associated DNA sequencing (RAD-seq) to find maximum likelihoods and have still been unsuccessful at confidently positioning *Aotus* within Cebidae (Valencia et al., 2018). The rapid diversification of Cebidae has made determining the exact phylogeny a difficult task that has required innovative methods to be applied, which have yet to yield confident results.

A platyrhine specific SINE element called Platy-1 is reported to have little activity in *Aotus*, virtually no current mobilization in squirrel monkey and capuchins (Jessica M. Storer et al., 2019), while marmoset exhibits extensive expansion of Platy-1 elements (Konkel et al., 2016). This could mean that a branch including *Aotus*, that later led to squirrel monkey and capuchins, diverged first, followed by the Callitrichinae branch that led to marmosets and tamarins and that the Platy-1 expansion took place after this split. This phylogeny was supported by Osterholz et al. (2009), using *Alu* elements. It could also mean that they emerged nearly simultaneously and the Platy-1 radiation in marmoset was simply delayed.

This study represents the most extensive use of *Alu* insertions to date in an attempt to resolve the placement of *Aotus* within the Cebidae family of platyrhine primates. The fact that we were unable to do so with confidence suggests that this issue may remain controversial into the foreseeable future.

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Appendix

APPENDIX 1

Feb. 20, 2019 NE-8 was replaced with NE-7: cj12-97, A02-737; another female marmoset
 Callimico 955 was replaced with our other callimico; 1504

New World Monkey DNA Panel			
DNA Plate: 150ul at 5ng/ul		V3: New plate format 2-20-19	
	1	2	3
A	TLE (negative control)	HeLa -Human	KB4544- Squirrel monkey
B	NS06006-Chimpanzee	African Green monkey	OR842 - Northern w-f saki
C	NG05356 - Woolly monkey	KB6701-Spider monkey	
D	NG05352-Spider monkey	OR749-Bolivian red howler monkey	
E	Callithrix jacchus- NE-7	OR690 - Pygmy marmoset	CRL1556- Owl monkey
F	1504- Callimico	NG05308-R-C must. tamarin	OR1522- Bolivian gray titi
G	OR621- Geoffroys s-b tamarin	CA003 - Capuchin	
H	30156 - Capuchin	30157 - Capuchin	

#	Species	Common name	Origin	ID
1	<i>Homo sapiens</i>	Human	ATCC	HeLa CCL-2
2	<i>Pan troglodytes</i>	Common Chimpanzee	IPBIR	NS06006
3	<i>Chlorocebus aethiops</i>	African Green monkey	ATCC	CCL70
4	<i>Lagothrix lagotricha</i>	Woolly monkey	Coriell	NG05356
5	<i>Ateles belzebuth</i>	White bellied spider monkey	SDFZ	KB6701
6	<i>Ateles geoffroyi</i>	black-handed spider monkey	Coriell	NG 05352
7	<i>Alouatta sara</i>	Bolivian red howler monkey	SDFZ	OR749
8	<i>Callithrix jacchus</i>	Common marmoset	NERPRC	cj12-97, A02-737
9	<i>Callithrix pygmea</i>	Pygmy marmoset	SDFZ	OR690
10	<i>Callimico goeldii</i>	Goeldi's marmoset	Alan Harris	1504
11	<i>Saguinus labiatus</i>	red-chested mustached tamarin	Coriell	NG05308
12	<i>Saguinus fuscicollis nigrifrons</i>	Geoffroys saddle-back tamarin	SDFZ	OR621
13	<i>Sapajus apella</i>	Tufted capuchin monkey	KP	CA003
14	<i>Sapajus apella</i>	Tufted capuchin monkey	KP	30156
15	<i>Sapajus apella</i>	Tufted capuchin monkey	KP	30157
16	<i>Saimiri s. sciureus</i>	Squirrel monkey	SDFZ	KB4544
17	<i>Aotus trivirgatus</i>	Owl monkey	ATCC	CRL1556
18	<i>Pithecia p. pithecia</i>	Northern white-faced saki	SDFZ	OR842
19	<i>Callicebus d. donacophilus</i>	Bolivian gray titi	SDFZ	OR1522

ATCC: From cell lines provided by the American Type Culture Collection
 IPBIR: Integrated Primate Biomaterials and Information Resource
 Coriell: Coriell Institute for Medical Research, 403 Haddon Avenue, Camden, NJ
 SDFZ: San Diego Frozen Zoo, Conservation and Research for Endangered Species (CRES)
 NERPRC: New England Regional Primate Research Center
 KP Kimberley Phillips, Trinity University

APPENDIX 2

		size	size	size	size
Primer Name	Primer Sequence	Marm	Owl	Cebus	Squirrel
CMS_M_24_0_marm-F	TGACATCCTTGAGCCCCATG				
CMS_M_24_0_marm-R	TAGTTTAGACAGGGGCCCTG	867	561	867	841
CMS_M_24_1_cebus-F	GACATCCTGAACCCCTATGC				
CMS_M_24_1_cebus-R	AGGGGCCCTGAACTAACTC	856	550	856	830
CMS_M_37_1_cebus-F	AGTCTGTGTGAACTTGCAACTC				
CMS_M_37_1_cebus-R	TGAGCACCTACAAAGTGTACAG	857	525	851	849
CMS_M_92_1_cebus-F	TGAGTTCTTGCAAGGCTGG				
CMS_M_92_1_cebus-R	AAGGGCCTCAGAGTTAGAGAG	826	501	851	837
CMS_M_118_3_SM-F	GACAGTGTGCGCTTGAGTATAG				
CMS_M_118_3_SM-R	ACCCACAAGCCTACCACAAC	883	585	885	894
CMS_C_5_3_SM-F	GGGAGATTCTGGAGGGTTTC				
CMS_C_5_3_SM-R	TGCAAGTAGGAGTGACTAGTCTC	819	509	863	832
CMS_C_8_0_cebus-F	GGAGTATCCCATCAAGATATCCC				
CMS_C_8_0_cebus-R	TGGTATTGTGTCAAGGAGTAGG	757	479	778	788
CMS_C_8_3_SM-R	GCAACATGTACTGGGGTCTAC	786	508	807	817
CMS_C_20_0_cebus-F	GCTAGAACAGGAAGCAGATC				
CMS_C_20_0_cebus-R	TTCCCCTAAGACTGGCTATCC	826	528	833	829
CMS_C_25_1_marm-F	TGAGTTGCTGCCTAGTGACAG				
CMS_C_25_1_marm-R	GTGTTAGGGTTGGGAGGAAAG	800	487	795	803
CMS_C_25_3_SM-F	TCAGCTGAAGACCTGAGTTG				
CMS_C_25_3_SM-R	GGTTAGGGTTGGGAGGAAAG	813	500	808	816
CMS_C_30_0_cebus-F	TGCAGGTCAGGAATTGAGAG				
CMS_C_30_0_cebus-R	TCACCTAGAATCTGGCCAATAC	782	478	799	784
CMS_C_39_1_marm-F	TCCAGCTGCCACTCATATGAC				
CMS_C_39_1_marm-R	TCAACTCCTCTGCCTCACTG	732	403	740	810
CMS_C_39_3_SM-F	CCAGACCAGGGGAAAGAAATG				
CMS_C_39_3_SM-R	TCTGCCAAACTCCTCTCTC	811	482	818	803
CMS_C_46_0_Cebus-F	GTCTGGTTGGTGGCTATGC				
CMS_C_46_0_Cebus-R	TCAGTGCCTGGCATGGTAG	747	444	738	752
CMS_C_47_0_Cebus-F	AGTTTACTTGATGACACAGTGG				
CMS_C_47_0_Cebus-R	TGCTCCTGATCCTGGGAAATC	825	493	800	818
CMS_C_52_0_Cebus-F	GCTCTGAACTGCATATGTACCTC				
CMS_C_52_0_Cebus-R	AGTTTAGCAGCCAACCAAGG	768	460	774	803
CMS_C_64_1_Marm-F	ATCAGCCCATAAACACTTCCC				
CMS_C_64_1_Marm-R	GAAGCCCTGCACGATAATGAG	750	469	780	787
CMS_C_64_3_squirrel-F	GATCAGCCCATAAGACTTCCC				
CMS_C_64_3_squirrel-R	AGAGCTGGCTTCACTGGTAG	777	496	807	814

		size	size	size	size
Primer Name	Primer Sequence	Marm	Owl	Cebus	Squirrel
CMS_C_76_0_Cebus-F	ACTCGCGGTTGGTATTGTG				
CMS_C_76_0_Cebus-R	TCCCTTGCCCTGAATAGAC	832	515	877	830
CMS_C_76_1_Marm-F	TGCACTCGTGGTTGGTATTG				
CMS_C_76_1_Marm-R	reverse the same as cebus	835	518	880	833
CMS_C_78_0_Cebus-F	TCAGCAAGATCATTCCAACC				
CMS_C_78_0_Cebus-R	ACCATGGCCAGTTAACCTAGAG	851	551	873	869
CMS_C_78_3_Squirrel-F	CCATGAACTGCTGTCCCCATC				
CMS_C_78_3_Squirrel-R	CCATGGCCGGTTAACCTAGAG	818	518	840	836
CMS_S_17_0_Saimiri-F	CCTTCATGATCTGGCCATCTC				
CMS_S_17_0_Saimiri-R	GGCACATCTGAGTTGACATTAC	790	459	777	791
CMS_S_48_0_Saimiri-F	CTTAGCTCAGACCTCTCAGATT				
CMS_S_48_0_Saimiri-R	TATCTGTCAGCATGGCATGG	738	391	734	729
CMS_S_48_1_Cebus-F	TCAGAGCTCTCAGATTCAAGGAG				
CMS_S_48_1_Cebus-R	GACCCTGGGAAGTAAAATCTCC	846	499	842	837
CMS_S_48_2_Marm-F	same forward as Cebus				
CMS_M_4_0_Marm-F	CAATCAGGAAATTCACTGAGGT				
CMS_M_4_0_Marm-R	TCCTAGCCACTCCTGATTAG	775	426	769	744
CMS_M_27_0_Marm-F	TCATGCAGCATTCTTACTCAG				
CMS_M_27_0_Marm-R	CCTAGTGACCATAGCACAGTCC	394	89	364	?
CMS_M_27_3_Squirrel-R	CCTAATGACAACAGCACAGTCC				400
CMS_C_32	TAGCACACCCATcaactaaac				
	tgtatGGGTAGAACATCTGCCAG	multiple matches; near duplication			
CMS_C_37_0_Cebus-F	CTGTGTACTGCCACAGACTGG				
CMS_C_37_0_Cebus-R	AACGTATGTAAGGCCATGGACA	546	242	555	553
CMS_C_58_0_Cebus-F	CATGAGGCCCTGTTGAATTG				
CMS_C_58_0_Cebus-R	GGGAAATTCTCTGTGACCTTG	881	563	904	868
CMS_C_58_2_Owl-R	CTGTGACCTTGTGAAATCTCC	871	553	894	858
CMS_C_63_0_Cebus-F	AAGTGAAGGTACAAGGAATTGG				
CMS_C_63_0_Cebus-R	TTACATTCAGCAGTGCTGG	661	325	625	632
CMS_C_72_0_Cebus-F	TTTCATAGTGTCCCCACAATC				
CMS_C_72_0_Cebus-R	GCAACAAACAATAGACCCCTGAGA	923	617	929	922
CMS_C_72_2_Owl-F	GTCCAGCAGTGTGAGTTCTCA	602	296	608	602
CMS_C_77_1_Marm-F	AACAATTATGCAGCATGTGAGC				
CMS_C_77_1_Marm-R	CTGGTCAGACTAGATAACCCACA	569	256	567	572

		size	size	size	size
Primer Name	Primer Sequence	Marm	Owl	Cebus	Squirrel
COS_C_5_3_Squirrel-Fa	GCCAGAGTTCACTTGACTT				
COS_C_5_3_Squirrel-R	CTGGGTGTTGCTCTTCCC	437	702	703	714
COS_C_5_0_Cebus-Fa	AAGATGAAGAACCGCACATGGA	380	645	646	657
COS_C_16_0_Cebus-F	GGTGTCTGTCAGATCTTGTCT				
COS_C_16_0_Cebus-R	AACAAGCCACAGATTGGAGAAT	142	446	460	450
COS_C_26_3_Squirrel-F	GCTGGAAAGGTTGAGTTCTG				
COS_C_26_3_Squirrel-R	CAACGTGGACACTGATAGGC	556	887	872	870
COS_C_28_2_Owl-F	AACACATGCCCTCCTCTAGAC				
COS_C_28_2_Owl-R	TCCTCAGTTATCCTCCACTCTG	544	880	876	859
COS_C_29_0_Cebus-F	ACTACGTGAGCCTAGCAGTTAG				
COS_C_29_0_Cebus-R	AGCAGAAGAACAGCAGCTTAG	496	825	805	802
COS_C_32_0_Cebus-F	GACATGGGCAGCCTATTGTTTC				
COS_C_32_0_Cebus-R	GGGAGGGAGATTGCTAACACTG	481	797	802	807
COS_C_32_3_Squirrel-F	GCAGCCTATTGTTCTCTACC				
COS_C_32_3_Squirrel-R	GCACGCATTAGGATCACTGAG	497	813	818	823
COS_C_35_3_Squirrel-F	GTATCTCTaGCACCTCACACAG				
COS_C_35_3_Squirrel-R	AACCTtCAGTAACCCCTCTG	424	751	775	752
COS_C_35_2_Owl-F	CCTTCTTGTATCTCTGGCACCT				
COS_C_35_2_Owl-R	CCACTTATGAGCAACTCTGTTCA	188	514	539	?
COS_C_36_3_Squirrel-F	TCCATACTGAGCTGCTGGAG				
COS_C_36_3_Squirrel-R	TCCTGGCCTGTTTATTACCTG	526	747	855	836
COS_C_41_3_Squirrel-F	ATGCCAGCTATGTTGCTAG				
COS_C_41_3_Squirrel-R	CTTGTGTGGTCCTGAAGC	517	836	829	842
COS_C_43_2_Owl-F	AGACACAGGAACCATGGTAGG				
COS_C_43_2_Owl-R	TCCAGGCAATGAGAGCACTAG	538	890	872	866
COS_C_51_2_Owl-F	CCATCTCATACGGGCAAAGATG				
COS_C_51_2_Owl-R	CTTCTGGGCAAAGTTCAGG	469	800	793	789
COS_C_51_3_Squirrel-F	CCATTTCATACGGGCAAAGATG				
COS_C_51_3_Squirrel-R	TGGGGCAAAGTTCAGGGTAG	465	796	789	785
COS_C_52_3_Squirrel-F	AGTAGAATGTCAAGGGCTCTG				
COS_C_52_3_Squirrel-R	CCCACTCCTGTTCTAATGACAG	471	883	878	885
COS_C_72_3_Squirrel-F	ACTGTGCTGCCTACTTGTCA				
COS_C_72_3_Squirrel-R	GTTGAGCAGAGCAGAGTTGTG	568	882	885	873
COS_C_74_0_Cebus-F	TTCAGGATTGTGGAGGTAGGG				
COS_C_74_0_Cebus-R	AAGGAAAGAGGGATGGAGGAG	506	812	832	810
COS_C_78_0_Cebus-F	CCAGTTCAACTTGGGGCTATG				
COS_C_78_0_Cebus-R	CTGTATTCTCAGCATCCACAAAC	520	827	825	833
COS_C_100_0_Cebus-F	AGTTATTACACTGGGCTCTGG				
COS_C_100_0_Cebus-R	GACTGCTCATACACTTGACCTG	531	844	838	855
COS_C_102_0_Cebus-F	GAGAGGGGAGAAATCGTTGTAG				
COS_C_102_0_Cebus-R	GGAATGTATGTGGGTCTCTAGC	583	891	888	892

		size	size	size	size
Primer Name	Primer Sequence	Marm	Owl	Cebus	Squirrel
COS_C_128_2_Owl-F	TTCTTGCTCTTGTGGAAGTGG				
COS_C_128_2_Owl-R	ATGTGCTGGCTAAAACAAGG	542	865	851	869
COS_O_61_3_Squirrel-F	GCTTGTCTCCTACTGCTCTTC				
COS_O_61_3_Squirrel-R	TCCTCAGGGCACTTACAGATG	528	840	840	847
COS_O_84_0_Owl-F	TGTTTGCAAGCAGGACATAGG				
COS_O_84_0_Owl-R	TTGTCAGGAGGGCTGGAAAC	555	868	869	869
COS_O_92_1_Cebus-F	GGGACACAGATCCTTGATGC				
COS_O_92_1_Cebus-R	AGGGTATACTGGGTGAACCTCC	542	833	845	880
COS_O_104_3_Squirrel-F	ACAAACAGCAGAGATATCCTAGGG				
COS_O_104_3_Squirrel-R	CAGGGAACAGGCCAGAAATTG	494	815	809	808
COS_O_159_3_Squirrel-F	GCACCAAGTCTCGTCTTAGG				
COS_O_159_3_Squirrel-R	TCATCATGACATGTGGTCCC	541	833	855	850
COS_O_208_1_Cebus-F	CAACACAGTACTGAAAGCCCTAG				
COS_O_208_1_Cebus-R	ACCTGGCCAACCTCAGAATGG	620	866	870	871
COS_S_42_0_Saimiri-F	CACTCTGCTAATCTCCATGGG				
COS_S_42_0_Saimiri-R	CAGCCCTCAGGGTACACACACATC	502	802	803	799
COS_C_33_0_Cebus-F	GTTAtAGCAACACAACGGGATG				
COS_C_33_0_Cebus-R	ATCACCAAGACACTGAAAGCAAA		583	590	
COS_C_33-marmoset-F	GGTTAcAGCAACAtAGCAGGA				
COS_C_33-marmoset-R	ATCACCAAGACACTGGAAGCA	267			570
COS_C_79_0_Cebus-F	TTGACTGCAAACTAGGTACAAAA	360	674	690	
COS_C_79_0_Cebus-R	CCCACAACTTCCCATTACCTT				
COS_C_79-marmoset-R	CAGCTCCCCATTACCTTCC	356	670		671
COS_C_3_0_Cebus-F	TTTCAACCAGCCTGAAGTAGTG			543	
COS_C_3_0_Cebus-R	CCATGTATCTGGAGAAGACATTG	209	522		541
COS_C_20_0_Cebus-F	AGCAGTGGCCTCAGTGAATA	132	461	474	
COS_C_20_0_Cebus-R	ACAAACCTGAACATGCACCA				
COS_C_20_marmoset-F	GCTACAAAGTAACACACAAATCAGC	154			492
COS_C_22_0_Cebus-F	TGGATAGGCATTACTGTGTTATT				
COS_C_22_0_Cebus-R	GGTACATTAAATCATGTCCTTGAG		799	752	
COS_C_22_marmoset-F	GGGCATTGCTTGTTCATTAATAC				
COS_C_22_marmoset-R	GTGCATTTCAATCATGTCCTTG	467			781
COS_C_25_0_Cebus-F	TCAAAACCTCCCCCTCAAACAC				
COS_C_25_0_Cebus-R	GGGGATGCAGTCAGTCTTTC		890	884	
COS_C_25_marmoset-R	TTGTCTAATGCTTGTCTGCATC	431			745
COS_C_37_0_Cebus-F	AGAGAGTAAGGCAGGGAAAGAA				
COS_C_37_0_Cebus-R	TTCCCTGCAGAACATTGCTATAC	485	530	810	788
COS_C_50_0_Cebus-F	CAGATTGAATACTGCTACCTTGT				
COS_C_50_0_Cebus-R	CTGAAATATCAGCAATTATGTTGAAG	170	x	478	471
COS_C_50_2_Owl-F	TGTTAAGTAAACTGCAGCCCTTC	147	462		x

		size	size	size	size
Primer Name	Primer Sequence	Marm	Owl	Cebus	Squirrel
COS_C_65_0_Cebus-F	GGCCACATAGATCAATTCTGA				
COS_C_65_0_Cebus-R	ACCTACATTCACAGTTGGATTT	386	692	691	694
COS_O_23_1_Cebus-F	GGAGGAAGATATTATTGGCTCAA				
COS_O_23_1_Cebus-R	gaCATTGCTGAATTCCCTCCTAA	292	603	601	604
COS_O_34_1_Cebus-F	CTCACAAACTCCTTCCAGTCC				
COS_O_34_1_Cebus-R	GCCTCACTGtggttcattt	409	746	697	751
COS_O_44_1_Cebus-F	AGTCACTGCCAACAGAGACTTC				
COS_O_44_1_Cebus-F	ATTGCATGTGGCATTCTTCTA	313	572	623	630
COS_O_59_1_Cebus-F	gAGATTGAAAAGCATAGCTTGAGA				
COS_O_59_1_Cebus-R	GCAATTATTCACCCCTTCA	172			
COS_O_59_0_Owl-R	CCCCTTACCGACATTCTGAAC	218	533	472	497
COS_S_4_1_Cebus-F	ACAGGTAAGGTTCAAGGaaagt				
COS_S_4_1_Cebus-R	GAAATTACCCCAGGAAgTTGG	344	643	645	644
COS_S_14_1_Cebus_F	TCAAAGTTTCAGGTCAATTGATT				
COS_S_14_1_Cebus_R	AATCTGCTCATTCCTTACATGAT	175	488	493	496

		size	size	size	size
Primer Name	Primer Sequence	Marm	Owl	Cebus	Squirrel
MOS_M_5_0_Marm-F	TCACTGGGAACTGGCTTAG				
MOS_M_5_0_Marm-R	AAGGGTGGCTGCAAATAATGG	881	880	564	883
MOS_M_5_2_Owl-F	same as Marm forward				
MOS_M_5_2_Owl-R	GAAGGGTGGCTGCAAATAATG	882	881	565	884
MOS_M_31_0_Marm-F	TAAGCCCCAGGAAATGTAGCC				
MOS_M_31_0_Marm-R	AGAGGCAAATGTGAACGTGAG	839	827	520	833
MOS_M_31_2_Owl-F	CTTAGTTGCTCGGTCCAAGTG				
MOS_M_31_2_Owl-R	CAGTGGACTTGGCTTACAGAC	No	849	540	855
MOS_M_31_3_Squirrel-F	GGAAGATCGGCCCTAACAAAC				
MOS_M_31_3_Squirrel-R	TGGCTTACAGACAGTCATTGC	878	865	No	871
MOS_M_35_2_Owl-F	AGACCCCAGCCAAATGAATTC				
MOS_M_35_2_Owl-R	CTATGTCTTGCTGTGGAGGAG	860	848	537	865
MOS_M_36_3_Squirrel-F	CGTTAGCAACCTTCCCTCAAG				
MOS_M_36_3_Squirrel-R	ATGCTTGGCCACTGTACTGAC	866/570	892	572	892
MOS_M_52_0_Marm-F	GAGCCTCCCAATTTCATC				
MOS_M_52_0_Marm-R	CATTGGTGTGTGGGGATG	822	841	517	830
MOS_M_63_3_Squirrel-F	ACTGTACCATGGCACAAACAG				
MOS_M_63_3_Squirrel-R	TGAGCTTACGGGGACTAAGG	827	828	504	830
MOS_M_72_2_Owl-F	ACTGGGGTGAAGACAAACTGG				
MOS_M_72_2_Owl-R	CAAAAGTTCTGTGTGCTTCC	774	782	469	774
MOS_O_3_2_Marm-F	CTTGCTTGGAAAAGTGAACC				
MOS_O_3_2_Marm-R	GGATTGGATGCCCTGACTTG	882	876	584	853
MOS_O_6_0_Owl-F	ACCTGGCTTGGAAACTAGTC				
MOS_O_6_0_Owl-R	CAAAGGTGCCAACTTCTCCTG	855	855	543	863
MOS_O_6_3_Squirrel-F	AGTCGTGTGAAGGGTAGTG				
MOS_O_6_3_Squirrel-R	GGGCACTCACATGTCACTAAC	No?	864	554	872
MOS_O_38_0_Owl-F	ACCACCATCATTGACTCGTTC				
MOS_O_38_0_Owl-R	TCCTGTCCTGTCAACTGAAG	736	744	422	744
MOS_O_38_2_Marm-F	ACACCCGCTTAATTGCTAACTG				
MOS_O_38_2_Marm-R	same reverse as owl	864	872	550	872
MOS_O_38_3_Squirrel-F	CATCATCATGGACTCGTCTGG				
MOS_O_38_3_Squirrel-R	ATCTCCTGTCCTGTCAACTG	737	745	423	745
MOS_O_50_0_Owl-F	AATACTGCACACGACACCTTG				
MOS_O_50_0_Owl-R	TGGTGCAGATATGGGTGAAC	No	773	469	769
MOS_O_50_2_Marm-F	TGCAAGCGTTCTGGAATCC				
MOS_O_50_2_Marm-R	CGATCAGAGTCCAGTCTAAC	741	738	434	734
MOS_O_50_3_Squirrel-F	same forward as owl				
MOS_O_50_3_Squirrel-R	ATGTTAGCCAGTTCAGGAAGC	876	874	570	870
MOS_O_58_0_Owl-F	GTGCTTTGAGTTGCCCTTG				
MOS_O_58_0_Owl-R	ATTGTAGTCTGCCAGCCAAG	843	844	539	861
MOS_O_59_2_Marm-F	CAAGTCCTCTGGCTCTCTTC				
MOS_O_59_2_Marm-R	ACCCTTGACTTCTGTGAGG	847	865	522	888
MOS_O_59_3_Squirrel-F	CAGTGCAGGATGAACATAGGC				
MOS_O_59_3_Squirrel-R	ATGATTCCGCCATGTTCACAG	849	867	524	890

		size	size	size	size
Primer Name	Primer Sequence	Marm	Owl	Cebus	Squirrel
CMO_M_29_1_Cebus-F	TTCTGCTGTGTTCTGGTTG				
CMO_M_29_1_Cebus-R	CACTGGAGAATTGGCAGTCC	845	869	858	525
CMO_M_69_2_Owl-F	GCAATTGTATGTGGACGAGAGG				
CMO_M_69_2_Owl-R	ATGAAGCCTCCCTGACCTC	863	874	868	549
CMO_O_45_2_Marm-F	TCCAAGAGCTTTGTGTGTG				
CMO_O_45_2_Marm-R	ACTGCGGACTGATGGGATTG	830	832	835	517
CMO_C_14_0_Cebus-F	CTCTGTGGAGTAGGGTCAGAAA				
CMO_C_14_0_Cebus-R	TCTCAGTCTCCATTGACAATACC	763	734	798	421
CMO_C_24_Squirrel-F	CTTGTGGACCCAATACCATCAT				
CMO_C_24_Squirrel-R	TAGAGCTCAGGACAAAGCATT	478	480	493	179
CMO_M_30_2_Owl-F	CTCCTTCCGCTTCATTCAATA				
CMO_M_30_2_Owl-R	TTTGTATCACACCCCACAAGGTT	571/709	573	586	291
CMO_O_45_0_Owl-F	CATTGCCAGAACAGAAAGCAGTC				
CMO_O_45_0_Owl-R	AAGCAAAGCTACTTAGAACTGGTC	677	677	680	362
CMO_O_31_0_Owl-F	TCTGAAACACAGAGTGTTCAGGT				
CMO_O_31_0_Owl-R	GACCTTGGCACAGAGACACA	475	484	451	131
CMO_O_54_2_Marm-F	GGGAGGTATTCAACTGAGACAGA				
CMO_O_54_2_Marm-R	GGCTACAAGTGTCCaaagtattaaa	898		949	
CMO_O_58_1_Cebus-F	ACCA GTT CCT GT TAG GC CT				
CMO_O_58_1_Cebus-R	GTGAATGGGTAGAACGCATTCC	541	542	438	238

APPENDIX 3

#	Species	ID	Locus-CMS									
			24-marm	24-cebus	37-cebus	92-cebus	118-SM	5-SM	8-cebus	8-SM	20-cebus	
1	<i>Homo sapiens</i>	HeLa CCL-2	0 0	0 0	0 0	-9 -9	-9 -9	0 0	0 0	0 0	0 0	-9 -9
2	<i>Pan troglodytes</i>	NS06006	-9 -9	-9 -9	0 0	-9 -9	-9 -9	-9 -9	0 0	0 0	0 0	-9 -9
3	<i>Chlorocebus aethiops</i>	CCL70	0 0	-9 -9	0 0	-9 -9	-9 -9	-9 -9	-9 -9	-9 -9	-9 -9	-9 -9
4	<i>Lagothrix lagotricha</i>	NG05356	0 0	-9 -9	1 1	1 1	0 0	1 1	1 1	1 1	1 1	0 0
5	<i>Ateles belzebuth</i>	KB6701	0 0	0 0	1 1	1 1	0 0	1 1	1 1	1 1	1 1	0 0
6	<i>Ateles geoffroyi</i>	NG 05352	0 0	0 0	1 1	1 1	0 0	1 1	1 1	1 1	1 1	-9 -9
7	<i>Alouatta sara</i>	OR749	0 0	0 0	0 1	1 -9	-9 0	0 0	1 1	1 1	1 1	0 0
8	<i>Callithrix jacchus</i>	cj12-97, A02-737	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1
9	<i>Callithrix pygmaea</i>	OR690	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1
10	<i>Callimico goeldii</i>	1504	1 1	1 1	1 1	1 1	1 1	-9 -9	1 1	1 1	1 1	1 1
11	<i>Saguinus labiatus</i>	NG05308	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1
12	<i>Saguinus fuscicollis nigrifrons</i>	OR621	1 1	-9 -9	1 1	1 1	1 1	1 1	-9 -9	1 1	1 1	1 1
13	<i>Sapajus apella</i>	CA003	1 1	1 1	1 1	1 1	1 1	0 0	1 1	1 1	1 1	1 1
14	<i>Sapajus apella</i>	30156	1 1	1 1	1 1	1 1	1 1	0 0	1 1	-9 -9	-9 -9	1 1
15	<i>Sapajus apella</i>	30157	1 1	1 1	1 1	1 1	1 1	0 0	1 1	1 1	1 1	1 1
16	<i>Saimiri s. sciureus</i>	KB4544	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1
17	<i>Aotus trivirgatus</i>	CRL1556	0 0	0 0	1 1	0 0	0 0	0 0	0 0	1 1	1 1	0 0
18	<i>Pithecia p. pithecia</i>	OR842	-9 -9	0 0	1 1	1 1	0 0	-9 -9	1 1	1 1	1 1	0 0
19	<i>Callicebus d. donacophilus</i>	OR1522	0 0	0 0	-9 -9	1 0	0 0	1 1	1 1	1 1	1 1	0 0

#	Species	ID	Locus-CMS									
			25-marm	25-SM	30-cebus	39-marm	39-SM	46-cebus	47-cebus	52-cebus	64-marm	
1	<i>Homo sapiens</i>	HeLa CCL-2	0 0	0 0	0 0	0 0	-9 -9	-9 -9	1 1	-9 -9	-9 -9	
2	<i>Pan troglodytes</i>	NS06006	0 0	0 0	0 0	0 0	-9 -9	-9 -9	0 0	-9 -9	-9 -9	
3	<i>Chlorocebus aethiops</i>	CCL70	0 0	0 0	0 0	0 0	-9 -9	-9 -9	-9 -9	0 0	0 0	-9 -9
4	<i>Lagothrix lagotricha</i>	NG05356	0 0	0 0	1 1	1 1	1 1	1 1	-9 -9	-9 -9	0 0	0 0
5	<i>Ateles belzebuth</i>	KB6701	0 0	0 0	1 1	1 1	1 1	-9 -9	-9 -9	1 1	0 0	0 0
6	<i>Ateles geoffroyi</i>	NG 05352	0 0	0 0	1 1	1 1	1 1	-9 -9	-9 -9	1 1	0 0	0 0
7	<i>Alouatta sara</i>	OR749	0 0	0 0	1 1	1 1	1 1	1 1	-9 -9	1 1	0 0	0 0
8	<i>Callithrix jacchus</i>	cj12-97, A02-737	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1
9	<i>Callithrix pygmea</i>	OR690	1 1	1 1	1 1	-9 -9	1 1	1 1	1 1	1 1	1 1	1 1
10	<i>Callimico goeldii</i>	1504	1 1	1 1	1 1	-9 -9	-9 -9	1 1	-9 -9	1 1	1 1	1 1
11	<i>Saguinus labiatus</i>	NG05308	-9 -9	-9 -9	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1
12	<i>Saguinus fuscicollis nigricans</i>	OR621	-9 -9	-9 -9	1 1	1 1	1 1	1 1	1 1	1 1	1 1	-9 -9
13	<i>Sapajus apella</i>	CA003	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1
14	<i>Sapajus apella</i>	30156	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1
15	<i>Sapajus apella</i>	30157	1 1	1 1	1 1	-9 -9	1 1	1 1	1 1	1 1	1 1	1 1
16	<i>Saimiri s. sciureus</i>	KB4544	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1
17	<i>Aotus trivirgatus</i>	CRL1556	0 0	0 0	1 1	0 0	0 0	0 0	0 0	0 0	0 0	0 0
18	<i>Pithecia p. pithecia</i>	OR842	0 0	0 0	0 0	0 0	1 1	1 1	0 0	0 0	0 0	-9 -9
19	<i>Callicebus d. donacophilus</i>	OR1522	0 0	0 0	0 0	0 0	1 1	1 1	0 0	0 0	0 0	-9 -9

#	Species	ID	Locus-CMS									
			64-SM	76-cebus	76-marm	78-cebus	78-SM	17-SM	48-SM	48-cebus	48-marm	
1	<i>Homo sapiens</i>	HeLa CCL-2	-9 -9	0 0	0 0	-9 -9	-9 -9	-9 -9	0 0	0 0	0 0	0 0
2	<i>Pan troglodytes</i>	NS06006	0 0	0 0	0 0	0 0	-9 -9	-9 -9	0 0	0 0	0 0	0 0
3	<i>Chlorocebus aethiops</i>	CCL70	0 0	0 0	0 0	0 0	-9 -9	-9 -9	0 0	0 0	0 0	0 0
4	<i>Lagothrix lagotricha</i>	NG05356	0 0	1 1	1 1	-9 -9	0 0	0 0	-9 -9	1 1	1 1	1 1
5	<i>Ateles belzebuth</i>	KB6701	0 0	1 1	1 1	0 0	0 0	0 0	-9 -9	1 1	1 1	1 1
6	<i>Ateles geoffroyi</i>	NG 05352	0 0	1 1	1 1	0 0	0 0	0 0	-9 -9	1 1	1 1	1 1
7	<i>Alouatta sara</i>	OR749	0 0	1 1	1 1	0 0	0 0	0 0	-9 -9	1 1	1 1	1 1
8	<i>Callithrix jacchus</i>	cj12-97, A02-737	1 1	1 1	1 1	1 1	-9 -9	-9 -9	-9 -9	1 1	1 1	1 1
9	<i>Callithrix pygmea</i>	OR690	1 1	1 1	1 1	1 1	1 1	1 1	-9 -9	1 1	1 1	1 1
10	<i>Callimico goeldii</i>	1504	1 1	1 1	1 1	1 1	1 1	1 1	-9 -9	-9 -9	1 1	-9 -9
11	<i>Saguinus labiatus</i>	NG05308	1 1	1 1	1 1	1 1	1 1	1 1	-9 -9	1 1	1 1	1 1
12	<i>Saguinus fuscicollis nigricans</i>	OR621	1 1	1 1	1 1	1 1	1 1	1 1	-9 -9	-9 -9	1 1	1 1
13	<i>Sapajus apella</i>	CA003	-9 -9	-9 -9	1 1	1 1	1 1	1 1	-9 -9	1 1	1 1	1 1
14	<i>Sapajus apella</i>	30156	-9 -9	-9 -9	1 1	1 1	1 1	1 1	-9 -9	1 1	1 1	-9 -9
15	<i>Sapajus apella</i>	30157	-9 -9	-9 -9	1 1	1 1	1 1	1 1	-9 -9	1 1	1 1	-9 -9
16	<i>Saimiri s. sciureus</i>	KB4544	1 1	1 1	1 1	-9 -9	1 1	1 1	-9 -9	1 1	1 1	1 1
17	<i>Aotus trivirgatus</i>	CRL1556	0 0	0 0	1 1	1 1	-9 -9	-9 -9	0 0	1 1	1 1	1 1
18	<i>Pithecia p. pithecia</i>	OR842	0 0	1 1	1 1	-9 -9	0 0	0 0	0 0	1 1	1 1	1 1
19	<i>Callicebus d. donacophilus</i>	OR1522	0 0	1 1	1 1	-9 -9	-9 -9	0 0	1 1	1 1	1 1	1 1

#	Species	ID	Locus-CMS									
			77-SM	77-marm	4-M	27-M	27-SM	37-C	58-C	58-O	63-C	72-C
1	<i>Homo sapiens</i>	HeLa CCL-2	0 0	0 0	-9 -9	-9 -9	-9 -9	0 0	-9 -9	-9 -9	-9 -9	-9 -9
2	<i>Pan troglodytes</i>	NS06006	0 0	0 0	-9 -9	-9 -9	-9 -9	0 0	0 0	-9 -9	-9 -9	-9 -9
3	<i>Chlorocebus aethiops</i>	CCL70	0 0	0 0	-9 -9	-9 -9	-9 -9	-9 -9	-9 -9	-9 -9	-9 -9	-9 -9
4	<i>Lagothrix lagotricha</i>	NG05356	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0	-9 -9
5	<i>Ateles belzebuth</i>	KB6701	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0	-9 -9
6	<i>Ateles geoffroyi</i>	NG 05352	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0	-9 -9
7	<i>Alouatta sara</i>	OR749	0 0	0 0	-9 -9	0 0	0 0	0 1	0 0	0 0	0 0	-9 -9
8	<i>Callithrix jacchus</i>	cj12-97, A02-737	1 1	1 1	1 1	1 1	1 1	1 1	1 1	-9 -9	-9 -9	-9 -9
9	<i>Callithrix pygmea</i>	OR690	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	-9 -9
10	<i>Callimico goeldii</i>	1504	-9 -9	-9 -9	-9 -9	-9 -9	-9 -9	-9 -9	-9 -9	-9 -9	-9 -9	-9 -9
11	<i>Saguinus labiatus</i>	NG05308	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1
12	<i>Saguinus fuscicollis nigricans</i>	OR621	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	-9 -9	1 1
13	<i>Sapajus apella</i>	CA003	-9 -9	-9 -9	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1
14	<i>Sapajus apella</i>	30156	-9 -9	-9 -9	1 1	1 1	1 1	1 1	-9 -9	1 1	1 1	-9 -9
15	<i>Sapajus apella</i>	30157	-9 -9	-9 -9	1 1	1 1	1 1	1 1	-9 -9	1 1	1 1	-9 -9
16	<i>Saimiri s. sciureus</i>	KB4544	1 1	1 1	1 1	-9 -9	1 1	1 1	-9 -9	1 1	1 1	-9 -9
17	<i>Aotus trivirgatus</i>	CRL1556	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 1	0 0	0 0
18	<i>Pithecia p. pithecia</i>	OR842	0 0	0 0	0 0	0 0	-9 -9	-9 -9	0 0	0 0	-9 -9	-9 -9
19	<i>Callicebus d. donacophilus</i>	OR1522	0 0	0 0	0 0	0 0	-9 -9	-9 -9	0 0	0 0	0 0	-9 -9

#	Species	ID	Locus-CMS										
			72-O	77-M	26-M	26-O	52-M	52-C	7-C	12-C	96-C	51-C	
1	<i>Homo sapiens</i>	HeLa CCL-2	-9 -9	0 0	-9 -9	-9 -9	-9 -9	0 0	0 0	0 0	0 0	-9 -9	-9 -9
2	<i>Pan troglodytes</i>	NS06006	0 0	0 0	-9 -9	-9 -9	-9 -9	0 0	0 0	0 0	0 0	-9 -9	-9 -9
3	<i>Chlorocebus aethiops</i>	CCL70	0 0	0 0	-9 -9	-9 -9	-9 -9	0 0	0 0	0 0	0 0	-9 -9	-9 -9
4	<i>Lagothrix lagotricha</i>	NG05356	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0	-9 -9
5	<i>Ateles belzebuth</i>	KB6701	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0	-9 -9	-9 -9
6	<i>Ateles geoffroyi</i>	NG 05352	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0	-9 -9	-9 -9
7	<i>Alouatta sara</i>	OR749	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0	-9 -9	-9 -9
8	<i>Callithrix jacchus</i>	cj12-97, A02-737	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 0	1 1	1 1	-9 -9
9	<i>Callithrix pygmea</i>	OR690	1 1	1 1	1 1	1 1	1 1	1 1	-9 -9	1 0	1 1	1 1	-9 -9
10	<i>Callimico goeldii</i>	1504	1 1	1 1	1 1	1 1	1 1	1 1	-9 -9	1 1	1 1	-9 -9	-9 -9
11	<i>Saguinus labiatus</i>	NG05308	1 1	-9 -9	1 1	-9 -9	1 1	-9 -9	1 1	-9 -9	1 0	1 1	-9 -9
12	<i>Saguinus fuscicollis nigrifrons</i>	OR621	1 1	-9 -9	1 1	-9 -9	1 1	-9 -9	1 1	1 1	1 0	1 1	-9 -9
13	<i>Sapajus apella</i>	CA003	1 1	1 1	1 1	1 1	1 1	-9 -9	1 1	1 1	0 0	1 1	-9 -9
14	<i>Sapajus apella</i>	30156	1 1	1 1	1 1	-9 -9	-9 -9	-9 -9	-9 -9	1 1	1 1	1 1	-9 -9
15	<i>Sapajus apella</i>	30157	1 1	1 1	1 1	-9 -9	-9 -9	-9 -9	-9 -9	1 1	1 1	1 1	-9 -9
16	<i>Saimiri s. sciureus</i>	KB4544	1 1	1 1	1 1	-9 -9	-9 -9	1 1	-9 -9	-9 -9	1 1	1 1	-9 -9
17	<i>Aotus trivirgatus</i>	CRL1556	0 0	0 0	0 0	0 0	-9 -9	0 0	-9 -9	-9 -9	0 0	0 0	-9 -9
18	<i>Pithecia p. pithecia</i>	OR842	0 0	-9 -9	-9 -9	-9 -9	-9 -9	0 0	-9 -9	-9 -9	0 0	0 0	-9 -9
19	<i>Callicebus d. donacophilus</i>	OR1522	0 0	0 0	0 0	-9 -9	-9 -9	0 0	-9 -9	0 0	0 0	-9 -9	-9 -9

#	Species	ID	Locus-COS										
			5-SM	5-cebus	16-cebus	26-SM	28-O	29-cebus	32-cebus	32-SM	35-SM		
1	<i>Homo sapiens</i>	HeLa CCL-2	-9 -9	-9 -9	-9 -9	-9 -9	0 0	-9 -9	0 0	-9 -9	0 0	-9 -9	0 0
2	<i>Pan troglodytes</i>	NS06006	-9 -9	-9 -9	-9 -9	-9 -9	0 0	0 0	-9 -9	0 0	-9 -9	0 0	0 0
3	<i>Chlorocebus aethiops</i>	CCL70	-9 -9	-9 -9	-9 -9	0 0	-9 -9	0 0	-9 -9	-9 -9	-9 -9	-9 -9	-9 -9
4	<i>Lagothrix lagotricha</i>	NG05356	1 1	1 1	1 1	0 0	1 1	0 0	0 0	1 1	1 1	1 1	-9 -9
5	<i>Ateles belzebuth</i>	KB6701	1 1	1 1	1 1	-9 -9	1 1	1 1	0 0	0 0	1 1	1 1	-9 -9
6	<i>Ateles geoffroyi</i>	NG 05352	1 1	1 1	1 1	-9 -9	1 1	1 1	0 0	0 0	1 1	1 1	-9 -9
7	<i>Alouatta sara</i>	OR749	1 1	1 1	1 1	0 0	1 1	0 0	0 0	0 0	1 1	-9 -9	1 1
8	<i>Callithrix jacchus</i>	cj12-97, A02-737	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0
9	<i>Callithrix pygmea</i>	OR690	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0	0 0	-9 -9	0 0
10	<i>Callimico goeldii</i>	1504	0 0	0 0	0 0	0 0	0 0	-9 -9	-9 -9	0 0	0 0	-9 -9	0 0
11	<i>Saguinus labiatus</i>	NG05308	1 1	1 1	1 1	0 0	0 0	0 0	0 0	0 0	0 0	-9 -9	0 0
12	<i>Saguinus fuscicollis nigrifrons</i>	OR621	1 1	1 1	1 1	0 0	0 0	0 0	0 0	0 0	0 0	-9 -9	0 0
13	<i>Sapajus apella</i>	CA003	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	-9 -9
14	<i>Sapajus apella</i>	30156	1 1	1 1	1 1	1 1	1 1	-9 -9	-9 -9	1 1	1 1	-9 -9	1 1
15	<i>Sapajus apella</i>	30157	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	-9 -9	1 1
16	<i>Saimiri s. sciureus</i>	KB4544	1 1	1 1	1 1	-9 -9	1 1	1 1	1 1	1 1	1 1	1 1	1 1
17	<i>Aotus trivirgatus</i>	CRL1556	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	1 1	-9 -9
18	<i>Pithecia p. pithecia</i>	OR842	1 1	1 1	1 1	-9 -9	1 1	1 1	0 0	1 1	-9 -9	-9 -9	0 0
19	<i>Callicebus d. donacophilus</i>	OR1522	1 1	1 1	1 1	-9 -9	1 1	1 1	0 0	1 1	1 1	-9 -9	0 0

#	Species	ID	Locus-COS											
			35-O	36-SM	41-SM	43-O	51-O	51-SM	52-SM	71-SM	74-cebus			
1	<i>Homo sapiens</i>	HeLa CCL-2	0	0	-9	-9	-9	0	0	0	0	-9	-9	
2	<i>Pan troglodytes</i>	NS06006	0	0	0	0	0	0	0	0	0	0	-9	-9
3	<i>Chlorocebus aethiops</i>	CCL70	0	0	0	0	1	1	0	-9	-9	0	-9	-9
4	<i>Lagothrix lagotricha</i>	NG05356	0	0	0	0	0	0	0	0	0	0	0	0
5	<i>Ateles belzebuth</i>	KB6701	0	0	0	0	0	0	0	0	0	0	0	0
6	<i>Ateles geoffroyi</i>	NG 05352	0	0	0	0	0	0	0	0	0	0	0	0
7	<i>Alouatta sara</i>	OR749	1	1	0	0	0	0	0	0	0	0	0	0
8	<i>Callithrix jacchus</i>	cj12-97, A02-737	0	0	0	0	0	0	0	0	0	0	0	0
9	<i>Callithrix pygmaea</i>	OR690	0	0	0	0	0	0	0	0	0	-9	-9	0
10	<i>Callimico goeldii</i>	1504	0	0	0	0	0	0	-9	-9	0	0	0	0
11	<i>Saguinus labiatus</i>	NG05308	0	0	0	0	0	-9	-9	0	0	0	0	0
12	<i>Saguinus fuscicollis nigricans</i>	OR621	0	0	0	0	-9	-9	0	0	0	-9	-9	0
13	<i>Sapajus apella</i>	CA003	1	1	1	1	1	1	1	1	1	1	1	1
14	<i>Sapajus apella</i>	30156	1	1	1	1	1	-9	-9	-9	1	1	-9	-9
15	<i>Sapajus apella</i>	30157	-9	-9	1	1	1	1	-9	-9	1	1	-9	-9
16	<i>Saimiri s. sciureus</i>	KB4544	-9	-9	-9	-9	1	1	1	1	1	1	1	1
17	<i>Aotus trivirgatus</i>	CRL1556	1	1	-9	-9	-9	1	1	-9	-9	1	1	1
18	<i>Pithecia p. pithecia</i>	OR842	0	0	0	0	0	-9	-9	0	0	0	1	1
19	<i>Callicebus d. donacophilus</i>	OR1522	0	0	0	0	0	0	0	0	0	-9	-9	0

#	Species	ID	Locus-COS										
			78-cebus	100-cebus	02-cebus	128-O	61-SM	84-O	92-cebus	104-SM	159-SM		
1	<i>Homo sapiens</i>	HeLa CCL-2	0	0	0	-9	-9	0	0	-9	-9	1	1
2	<i>Pan troglodytes</i>	NS06006	0	0	0	-9	-9	0	0	-9	-9	1	1
3	<i>Chlorocebus aethiops</i>	CCL70	0	0	0	0	-9	-9	-9	-9	0	1	1
4	<i>Lagothrix lagotricha</i>	NG05356	1	1	0	0	0	1	1	-9	-9	0	1
5	<i>Ateles belzebuth</i>	KB6701	1	1	-9	-9	0	0	1	1	-9	-9	0
6	<i>Ateles geoffroyi</i>	NG 05352	1	1	0	0	0	1	1	-9	-9	0	0
7	<i>Alouatta sara</i>	OR749	1	1	0	0	0	1	1	0	0	1	0
8	<i>Callithrix jacchus</i>	cj12-97, A02-737	0	0	0	0	0	0	0	0	0	0	0
9	<i>Callithrix pygmaea</i>	OR690	0	0	0	0	0	0	0	0	0	0	0
10	<i>Callimico goeldii</i>	1504	-9	-9	0	0	0	-9	-9	0	-9	-9	0
11	<i>Saguinus labiatus</i>	NG05308	0	0	0	0	-9	-9	-9	0	0	0	-9
12	<i>Saguinus fuscicollis nigricans</i>	OR621	0	0	0	0	0	1	1	-9	-9	0	-9
13	<i>Sapajus apella</i>	CA003	1	1	1	1	1	1	1	-9	-9	1	1
14	<i>Sapajus apella</i>	30156	1	1	1	1	1	-9	-9	-9	-9	1	1
15	<i>Sapajus apella</i>	30157	1	1	1	1	1	1	-9	-9	1	1	1
16	<i>Saimiri s. sciureus</i>	KB4544	1	1	-9	-9	1	1	1	-9	-9	1	1
17	<i>Aotus trivirgatus</i>	CRL1556	1	1	1	1	1	1	-9	-9	1	1	1
18	<i>Pithecia p. pithecia</i>	OR842	1	1	-9	-9	0	0	-9	-9	0	0	1
19	<i>Callicebus d. donacophilus</i>	OR1522	-9	-9	-9	-9	0	0	1	1	-9	0	0

#	Species	ID	Locus-COS										
			208-cebu	42-SM	33-cebus	33-M	79-cebus	79-M	3-cebus	20-cebus	20-M		
1	<i>Homo sapiens</i>	HeLa CCL-2	0	0	0	-9	-9	-9	-9	0	0	-9	-9
2	<i>Pan troglodytes</i>	NS06006	0	0	0	-9	-9	-9	-9	0	0	0	-9
3	<i>Chlorocebus aethiops</i>	CCL70	0	0	0	-9	-9	-9	-9	0	0	0	-9
4	<i>Lagothrix lagotricha</i>	NG05356	1	1	0	-9	-9	-9	0	0	0	1	1
5	<i>Ateles belzebuth</i>	KB6701	1	1	0	-9	-9	-9	0	0	0	1	1
6	<i>Ateles geoffroyi</i>	NG 05352	1	1	0	-9	-9	-9	0	0	0	1	1
7	<i>Alouatta sara</i>	OR749	1	1	0	0	0	0	-9	-9	0	1	1
8	<i>Callithrix jacchus</i>	cj12-97, A02-737	0	0	0	0	0	0	0	0	0	0	0
9	<i>Callithrix pygmaea</i>	OR690	0	0	0	0	0	0	0	0	0	0	0
10	<i>Callimico goeldii</i>	1504	0	0	0	0	-9	-9	-9	0	0	0	0
11	<i>Saguinus labiatus</i>	NG05308	1	1	1	0	-9	-9	0	0	0	0	0
12	<i>Saguinus fuscicollis nigricans</i>	OR621	1	1	1	0	-9	-9	0	0	0	0	0
13	<i>Sapajus apella</i>	CA003	1	1	1	1	-9	-9	1	1	-9	1	1
14	<i>Sapajus apella</i>	30156	-9	-9	1	1	-9	-9	-9	-9	1	1	1
15	<i>Sapajus apella</i>	30157	-9	-9	1	1	-9	-9	-9	-9	1	1	1
16	<i>Saimiri s. sciureus</i>	KB4544	-9	-9	1	1	-9	-9	-9	-9	1	1	1
17	<i>Aotus trivirgatus</i>	CRL1556	-9	-9	1	1	-9	-9	1	1	1	1	1
18	<i>Pithecia p. pithecia</i>	OR842	-9	-9	0	0	-9	-9	-9	-9	1	1	-9
19	<i>Callicebus d. donacophilus</i>	OR1522	-9	-9	0	0	-9	-9	-9	-9	-9	0	0

#	Species	ID	Locus-COS									
			22-cebus	22-M	25-cebus	25-M	37-cebus	50-cebus	50-O	65-cebus	23-cebus	
1	<i>Homo sapiens</i>	HeLa CCL-2	-9	-9	-9	-9	0	0	-9	-9	-9	-9
2	<i>Pan troglodytes</i>	NS06006	-9	-9	-9	-9	0	0	-9	-9	-9	-9
3	<i>Chlorocebus aethiops</i>	CCL70	-9	-9	-9	-9	0	0	-9	-9	-9	-9
4	<i>Lagothrix lagotricha</i>	NG05356	-9	-9	-9	-9	1	1	1	0	-9	-9
5	<i>Ateles belzebuth</i>	KB6701	-9	-9	-9	-9	1	1	1	0	1	1
6	<i>Ateles geoffroyi</i>	NG 05352	0	0	-9	-9	1	1	1	0	0	0
7	<i>Alouatta sara</i>	OR749	0	0	-9	-9	1	1	1	-9	-9	1
8	<i>Callithrix jacchus</i>	cj12-97, A02-737	-9	-9	0	0	-9	-9	0	0	-9	-9
9	<i>Callithrix pygmea</i>	OR690	-9	-9	0	0	-9	-9	0	0	-9	-9
10	<i>Callimico goeldii</i>	1504	-9	-9	-9	-9	-9	-9	0	0	-9	-9
11	<i>Saguinus labiatus</i>	NG05308	0	0	0	0	0	0	-9	-9	0	0
12	<i>Saguinus fuscicollis nigrifrons</i>	OR621	0	0	0	0	0	0	-9	-9	0	0
13	<i>Sapajus apella</i>	CA003	1	1	-9	-9	1	1	1	-9	1	1
14	<i>Sapajus apella</i>	30156	1	1	-9	-9	1	1	1	-9	1	1
15	<i>Sapajus apella</i>	30157	1	1	-9	-9	1	1	1	-9	1	1
16	<i>Saimiri s. sciureus</i>	KB4544	-9	-9	-9	-9	1	1	1	1	-9	-9
17	<i>Aotus trivirgatus</i>	CRL1556	1	1	1	1	1	1	-9	-9	1	1
18	<i>Pithecia p. pithecia</i>	OR842	0	0	0	0	0	0	0	-9	-9	1
19	<i>Callicebus d. donacophilus</i>	OR1522	-9	-9	-9	-9	-9	-9	-9	-9	1	1

#	Species	ID	Locus-COS							
			34-cebus	44-cebus	59-cebus	59-O	4-cebus	14-cebus		
1	<i>Homo sapiens</i>	HeLa CCL-2	0	0	-9	-9	-9	-9	0	0
2	<i>Pan troglodytes</i>	NS06006	0	0	-9	-9	-9	-9	0	0
3	<i>Chlorocebus aethiops</i>	CCL70	0	0	-9	-9	-9	-9	0	0
4	<i>Lagothrix lagotricha</i>	NG05356	0	0	0	0	0	-9	0	0
5	<i>Ateles belzebuth</i>	KB6701	0	0	0	0	0	-9	0	-9
6	<i>Ateles geoffroyi</i>	NG 05352	0	0	0	0	0	-9	0	-9
7	<i>Alouatta sara</i>	OR749	0	0	-9	-9	-9	0	0	-9
8	<i>Callithrix jacchus</i>	cj12-97, A02-737	0	0	0	0	0	0	0	0
9	<i>Callithrix pygmea</i>	OR690	0	0	0	0	0	-9	0	0
10	<i>Callimico goeldii</i>	1504	0	0	-9	-9	-9	-9	0	-9
11	<i>Saguinus labiatus</i>	NG05308	0	0	0	-9	-9	0	0	0
12	<i>Saguinus fuscicollis nigrifrons</i>	OR621	0	0	0	-9	-9	0	0	0
13	<i>Sapajus apella</i>	CA003	1	1	1	1	1	1	1	1
14	<i>Sapajus apella</i>	30156	-9	-9	1	1	-9	-9	-9	1
15	<i>Sapajus apella</i>	30157	1	1	1	1	1	1	1	1
16	<i>Saimiri s. sciureus</i>	KB4544	1	1	1	1	1	1	1	1
17	<i>Aotus trivirgatus</i>	CRL1556	1	1	1	1	1	1	1	1
18	<i>Pithecia p. pithecia</i>	OR842	0	0	1	1	0	0	-9	0
19	<i>Callicebus d. donacophilus</i>	OR1522	0	0	1	1	0	0	0	-9

#	Species	ID	Locus-MOS									
			5-M	5-O	31-M	31-O	31-SM	35-O	36-SM	52-M	63-SM	
1	<i>Homo sapiens</i>	HeLa CCL-2	1	1	-9	-9	0	0	-9	-9	-9	-9
2	<i>Pan troglodytes</i>	NS06006	0	0	0	0	0	0	-9	-9	-9	-9
3	<i>Chlorocebus aethiops</i>	CCL70	0	0	0	0	0	-9	-9	-9	-9	-9
4	<i>Lagothrix lagotricha</i>	NG05356	0	0	0	0	0	0	0	0	-9	-9
5	<i>Ateles belzebuth</i>	KB6701	0	0	0	0	0	0	0	-9	-9	0
6	<i>Ateles geoffroyi</i>	NG 05352	0	0	0	0	0	0	0	-9	-9	0
7	<i>Alouatta sara</i>	OR749	0	0	0	0	0	0	0	0	-9	-9
8	<i>Callithrix jacchus</i>	cj12-97, A02-737	1	1	1	1	1	1	1	1	0	-9
9	<i>Callithrix pygmea</i>	OR690	1	1	1	1	1	1	1	1	0	-9
10	<i>Callimico goeldii</i>	1504	-9	-9	1	1	1	-9	1	1	0	-9
11	<i>Saguinus labiatus</i>	NG05308	1	1	1	1	1	-9	1	1	0	-9
12	<i>Saguinus fuscicollis nigricans</i>	OR621	1	1	1	1	1	1	1	1	0	-9
13	<i>Sapajus apella</i>	CA003	0	0	0	0	0	0	-9	-9	0	-9
14	<i>Sapajus apella</i>	30156	0	0	0	0	0	0	-9	-9	0	-9
15	<i>Sapajus apella</i>	30157	0	0	0	0	0	0	-9	-9	0	-9
16	<i>Saimiri s. sciureus</i>	KB4544	1	1	1	1	1	1	1	1	1	1
17	<i>Aotus trivirgatus</i>	CRL1556	1	1	1	1	1	1	1	1	1	1
18	<i>Pithecia p. pithecia</i>	OR842	0	0	0	0	0	-9	-9	0	0	-9
19	<i>Callicebus d. donacophilus</i>	OR1522	0	0	0	0	0	0	0	0	-9	0

#	Species	ID	Locus-MOS									
			72-O	3-M	6-O	6-SM	38-O	38-M	38-SM	50-O	50-M	
1	<i>Homo sapiens</i>	HeLa CCL-2	0	0	0	0	0	-9	-9	0	0	0
2	<i>Pan troglodytes</i>	NS06006	0	0	0	0	0	-9	-9	0	0	0
3	<i>Chlorocebus aethiops</i>	CCL70	0	0	-9	-9	0	0	-9	-9	0	0
4	<i>Lagothrix lagotricha</i>	NG05356	1	1	-9	-9	0	0	-9	-9	1	1
5	<i>Ateles belzebuth</i>	KB6701	1	1	1	1	0	0	0	0	1	1
6	<i>Ateles geoffroyi</i>	NG 05352	1	1	1	1	0	0	0	-9	1	1
7	<i>Alouatta sara</i>	OR749	-9	-9	1	1	-9	0	0	-9	1	1
8	<i>Callithrix jacchus</i>	cj12-97, A02-737	1	1	1	1	1	1	1	1	1	1
9	<i>Callithrix pygmea</i>	OR690	-9	-9	1	1	1	1	-9	-9	1	1
10	<i>Callimico goeldii</i>	1504	-9	-9	1	1	1	-9	-9	1	1	1
11	<i>Saguinus labiatus</i>	NG05308	1	1	1	1	-9	-9	-9	1	1	-9
12	<i>Saguinus fuscicollis nigricans</i>	OR621	-9	-9	1	1	-9	-9	-9	-9	1	1
13	<i>Sapajus apella</i>	CA003	1	1	1	1	0	0	0	0	0	0
14	<i>Sapajus apella</i>	30156	-9	-9	1	1	1	0	0	-9	-9	0
15	<i>Sapajus apella</i>	30157	-9	-9	-9	-9	0	0	0	-9	0	0
16	<i>Saimiri s. sciureus</i>	KB4544	1	1	1	1	1	1	1	1	1	1
17	<i>Aotus trivirgatus</i>	CRL1556	1	1	1	1	1	1	1	1	1	1
18	<i>Pithecia p. pithecia</i>	OR842	-9	-9	-9	-9	0	0	0	-9	1	1
19	<i>Callicebus d. donacophilus</i>	OR1522	-9	-9	1	1	0	0	0	0	1	1

#	Species	ID	Locus-MOS									
			50-SM	58-O	59-M	59-SM	26-M	26-O	21-M	21-C	21-SM	77-M
1	<i>Homo sapiens</i>	HeLa CCL-2	-9	-9	0	0	0	0	-9	-9	0	0
2	<i>Pan troglodytes</i>	NS06006	-9	-9	0	0	0	0	-9	-9	-9	0
3	<i>Chlorocebus aethiops</i>	CCL70	-9	-9	0	1	1	1	-9	-9	0	-9
4	<i>Lagothrix lagotricha</i>	NG05356	1	1	1	1	1	1	-9	-9	1	1
5	<i>Ateles belzebuth</i>	KB6701	1	1	1	1	1	1	0	-9	1	1
6	<i>Ateles geoffroyi</i>	NG 05352	1	1	1	1	1	1	0	-9	1	1
7	<i>Alouatta sara</i>	OR749	1	1	-9	1	1	1	-9	-9	1	1
8	<i>Callithrix jacchus</i>	cj12-97, A02-737	1	1	1	1	1	1	-9	-9	1	1
9	<i>Callithrix pygmea</i>	OR690	1	1	1	1	1	1	-9	-9	1	1
10	<i>Callimico goeldii</i>	1504	-9	-9	-9	-9	-9	-9	-9	1	1	-9
11	<i>Saguinus labiatus</i>	NG05308	-9	-9	1	1	1	1	-9	-9	1	1
12	<i>Saguinus fuscicollis nigricans</i>	OR621	1	1	1	1	1	1	-9	-9	1	1
13	<i>Sapajus apella</i>	CA003	0	0	1	1	1	1	-9	-9	0	-9
14	<i>Sapajus apella</i>	30156	0	0	-9	-9	1	1	-9	-9	0	-9
15	<i>Sapajus apella</i>	30157	0	0	-9	-9	1	1	-9	-9	0	-9
16	<i>Saimiri s. sciureus</i>	KB4544	1	1	-9	1	1	1	-9	-9	-9	1
17	<i>Aotus trivirgatus</i>	CRL1556	1	1	1	1	1	1	-9	-9	1	1
18	<i>Pithecia p. pithecia</i>	OR842	1	1	-9	-9	1	1	-9	-9	0	1
19	<i>Callicebus d. donacophilus</i>	OR1522	1	1	1	1	1	1	-9	-9	0	-9

#	Species	ID	Locus-CMO										
			29-C	69-O	45-M	14-C	24-SM	30-O	45-O	31-O	54-M	58-C	
1	<i>Homo sapiens</i>	HeLa CCL-2	0 0 0 0 0 0 -9 -9 -9 -9 -9 -9 -9 -9										
2	<i>Pan troglodytes</i>	NS06006	0 0 0 0 0 0 -9 -9 -9 -9 -9 -9 -9 -9										
3	<i>Chlorocebus aethiops</i>	CCL70	0 0 -9 -9 0 0 -9 -9 -9 -9 -9 -9 -9 -9										
4	<i>Lagothrix lagotricha</i>	NG05356	0 0 1 1 0 0 0 0 0 1 1 0 0 0										
5	<i>Ateles belzebuth</i>	KB6701	0 0 -9 -9 0 0 0 0 0 1 1 0 0 0										
6	<i>Ateles geoffroyi</i>	NG 05352	0 0 1 1 0 0 0 0 0 1 1 0 0 0										
7	<i>Alouatta sara</i>	OR749	0 0 -9 -9 0 0 -9 -9 0 0 1 1 0 0										
8	<i>Callithrix jacchus</i>	cj12-97, A02-737		1 1 1 1 1 1 -9 -9 1 1 1 1 1 1									
9	<i>Callithrix pygmea</i>	OR690	1 1 1 1 1 1 1 1 -9 -9 1 1 1 1										
10	<i>Callimico goeldii</i>	1504		1 1 -9 -9 1 1 -9 -9 1 1 1 1 1 1									
11	<i>Saguinus labiatus</i>	NG05308	1 1 -9 -9 1 1 -9 -9 1 1 1 1 1 1										
12	<i>Saguinus fuscicollis nigrifrons</i>	OR621	1 1 -9 -9 1 1 -9 -9 -9 -9 1 1 1 1										
13	<i>Sapajus apella</i>	CA003	1 1 -9 -9 1 1 1 1 1 1 -9 -9 1 1										
14	<i>Sapajus apella</i>	30156	1 1 -9 -9 -9 -9 1 1 1 1 -9 -9 1 1										
15	<i>Sapajus apella</i>	30157	1 1 -9 -9 -9 -9 1 1 1 1 -9 -9 1 1										
16	<i>Saimiri s. sciureus</i>	KB4544	0 0 0 0 0 0 0 0 0 1 1 0 0 0										
17	<i>Aotus trivirgatus</i>	CRL1556	1 1 -9 -9 -9 -9 1 1 1 1 1 1 1 1										
18	<i>Pithecia p. pithecia</i>	OR842	0 0 -9 -9 -9 -9 0 0 0 0 -9 -9 0 0										
19	<i>Callicebus d. donacophilus</i>	OR1522	0 0 -9 -9 0 0 -9 -9 0 0 -9 -9 0 0										