

# Ancient mtDNA Implies a Nonconstant Molecular Clock in the Human Holobaramin

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## Abstract

An analysis of 61 human complete mitochondrial genomes (six Neandertal, two Denisovan, and 52 modern *Homo sapiens*) revealed real differences well outside of the range modern *Homo sapiens* mtDNA diversity. Neandertals and *Homo sapiens* differed by an average of 190.9 single nucleotide differences (SNDs), with 104 fixed differences. Denisovans and *Homo sapiens* differed by an average of 370.8 SNDs, with 265 fixed differences. Denisovans and Neandertals differed by an average of 355 SNDs, with 349 fixed differences. When analyzed under the assumption of a molecular clock, results indicated that molecular substitution rates in human mtDNA must have been much higher at some point in the first 2000 years of earth history.

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## Introduction

Since the 1987 proposal of the mitochondrial Eve hypothesis (Cann et al. 1987), creationists have been keenly interested in reconciling human mitochondrial DNA with a young-earth chronology. According to the Eve hypothesis, the ancestor of modern human mitochondrial DNA can be traced to a single mtDNA type from Africa around 150,000 years ago by conventional dating. Early controversies about both the date and source of the mitochondrial ancestor (e.g., Templeton 1992; Maddison et al. 1992) have largely been put to rest with refinements in both data and methodology. For example, the use of median networks has greatly added to our ability to study geographic origins (e.g., Bandelt et al. 1995), and likelihood and coalescent methods have replaced earlier reliance on parsimony (e.g., Disotell 1999). Likewise, the ability to rapidly and accurately sequence entire mitochondrial genomes has dramatically improved the data analyzed (e.g., Ingman et al. 2000). Today, the recency of modern human ancestry and African origin are generally accepted among molecular anthropologists.

Creationist response to the mitochondrial Eve hypothesis has been mixed. Some creationists have suggested serious flaws in human mitochondrial DNA analyses (Lubenow 1994, 2004), but a much greater number of creationists have sought to somehow reconcile mitochondrial Eve with the historical Eve. For example,

Wieland (1998) emphasized that although human mitochondrial DNA was not proof of the biblical account of Adam and Eve, it was “**consistent** with it.” More recently, Carter (2007) published a human mtDNA consensus sequence, which he subsequently equated with the historical Eve’s actual mtDNA sequence (Carter et al. 2008). In contrast, Wood (2008) argued that the most recent common ancestor of modern *Homo sapiens* mitochondrial DNA could not be the historical Eve but instead would be one of Noah’s daughters-in-law.

Neandertals have long been recognized as human by creationists (Nelson 1948; Cuozzo 1998; Hartwig-Scherer 1998; Lubenow 2004; Wise 2005), and since 1997 numerous fragments of Neandertal DNA have been sequenced and analyzed (Krings et al. 1997; Krings et al. 1999; Krings et al. 2000; Ovchinnikov et al. 2000; Schmitz et al. 2002; Caramelli et al. 2006; Lalueza-Fox et al. 2006; Orlando et al. 2006; Krause et al. 2007; Lalueza-Fox et al. 2007; Green et al. 2008; Lalueza-Fox et al. 2008; Briggs et al. 2009; Lari et al. 2010; Lalueza-Fox et al. 2011), culminating in the publication of a draft Neandertal genome in 2010 (Green et al. 2010). More recently, a hominin fossil from the Denisova Cave in Siberia yielded mtDNA and nuclear genome sequences strikingly different from Neandertal and modern *Homo sapiens* sequences (Krause et al. 2010b; Reich et al. 2010). These studies have consistently shown that Neandertals and the Denisovan sequences are diagnosably different from modern humans, even when rate

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variation is taken into account (Gutiérrez et al. 2002).

The Neandertal and Denisovan nuclear genome sequences have also revealed low levels of introgression into the gene pool of modern *Homo sapiens*. In the case of the Neandertals, 1-4% of non-African polymorphisms appear to be of Neandertal origin (Green et al. 2010). Similarly, Reich et al. (2010) found evidence of interbreeding between the Denisovans and the ancestors of modern Melanesians. Following Marsh's (1947) interbreeding criterion for identifying members of the same baramin, this indirect evidence of interbreeding between these hominins suggests that modern humans, Neandertals, and Denisovans are all members of the same human baramin and therefore descendants of Adam and Eve. Since hominin fossils are post-Flood (Wood 2010) and Neandertals and Denisovans are geographically distant from the Ararat region, they are most likely post-Babel populations. By using dates inferred from Scripture (Genesis 5 and 11) and making the normal assumptions of the molecular clock hypothesis, we can test the molecular clock and infer a possible genetic history of post-Flood humans.

Especially useful in this regard is the recent publication of a *Homo sapiens* mtDNA genome sequenced from a fossil from Kostenki, Russia that is roughly contemporaneous with the Denisovan and most Neandertal fossils from which mtDNA genome sequences have been published (Krause et al. 2010a). The Denisovan fossils have been dated to 30-48 Kyr ago (Krause et al. 2010b), and four of the six Neandertal mtDNA genomes come from fossils dated to approximately 39-40 Kyr ago (Briggs et al. 2009). The Kostenki mtDNA genome came from a fossil dated to approximately 30 Kyr ago. These dates are obviously incompatible with a biblical chronology inferred from Genesis 5 and 11, but the relative dating of these fossils support the inference that they may have been contemporaries. Thus, the Kostenki mtDNA genome gives us insight into the genetic state of anatomically modern humans soon after the Flood, while Denisovans and Neandertals were still alive.

Before evaluating these sequences, though, we must address the question of sequencing accuracy. Carter (2009) and Criswell (2009) both remain skeptical of ancient DNA recovery and sequencing methods. Criswell concluded that published Neandertal DNA sequences suffered from post-mortem degeneration and that "the relationship of Neandertals to modern humans cannot be fully assessed using currently available Neandertal DNA sequences." These concerns were somewhat justifiable at the time they were written, but the present situation suggests that natural degeneration is unlikely to bias evaluation of Neandertal or Denisovan mtDNA genomes. There are now six Neandertal mtDNA genomes published, and they are more similar to each other than some modern human sequences are to each other (Briggs et al. 2010). Likewise, the two Denisovan mtDNA genomes are nearly identical (Reich et al. 2010). If random degeneration created artificial differences from modern human sequences, we should expect to see similar differences between independent ancient DNA samples. Instead, we find a remarkable similarity between independent ancient samples, suggesting that random degeneration is not a significant problem.

Given the likely accuracy of these ancient sequences, the complete mtDNA genomes of six Neandertals, two Denisovans, and the Kostenki *Homo sapiens* fossil can be re-examined from

a creationist perspective. Of particular interest is whether the Neandertals and Denisovans represent genetically distinct groups or merely variations of modern *Homo sapiens*. Carter (2009) implies that there are important differences between Neandertals and modern humans, but DeWitt and Skinner's (2001) analysis suggested that Neandertals "cannot be excluded from the range of human variability." If there are substantial differences as the conventional literature affirms (e.g., Krause et al. 2010b), then what do the Denisovan, Neandertal, and modern *Homo sapiens* sequences tell us about the rate of mtDNA substitutions prior to and immediately after the Flood?

## Methods

Complete mtDNA sequences from two Denisovans (FN673705 and FR695060), six Neandertals (NC\_011137, FM865407, FM865408, FM865409, FM865410, FM865411), and the Kostenki fossil (FN600416) were obtained from GenBank. As references for comparison and phylogenetic analysis, 53 *Homo sapiens* mtDNA genomes from a study by Ingman et al. (2000) were included in the analysis, as well as mtDNA genomes from two chimps (NC\_001643 and X93335), a bonobo (NC\_001644), and a gorilla (NC\_001645). Carter's (2007) Eve 1.0 consensus sequence was also included. Altogether, there were 67 sequences in the full dataset: four from non-human apes, two from Denisovans, six from Neandertals, 54 from *Homo sapiens*, and one artificial consensus sequence of modern *Homo sapiens*.

Two sequence alignments were generated using CLUSTALW (Thompson et al., 1994). The first alignment included all 67 mtDNA genomes in the full dataset and consisted of 16,606 aligned positions. The second alignment consisted of eight mtDNA sequences obtained from fossils that were putative contemporaries (the sequence of the Mezmaiskaya fossil was excluded since it was dated as significantly older than the other Neandertal sequences). This second alignment of ancient sequences was used for ancestral dating inferences and consisted of 16,577 aligned positions. All phylogenetic analysis on both alignments was conducted in MEGA 5 (megasoftware.net; Kumar et al. 2008).

For maximum-likelihood phylogenetic analysis, the best substitution model was selected using MEGA's model selection function. Twenty-four different models were examined based on six basic substitution models: General Time Reversible, Tamura-Nei, Tamura 3-parameter, Kimura 2-parameter, Jukes-Cantor, and Hasegawa-Kishinano-Yano. For each model, rate heterogeneity was modeled using a gamma distribution of rates, by including invariant sites, or both. Maximum likelihood values were calculated for each model, based on the fit of the data to a neighbor-joining tree, and the model with the lowest Bayesian Information Criterion score was used in phylogenetic analyses.

For the smaller dataset of ancient sequences, a simple clock calibration was performed based on the assumption that the date of death of the individual taxa was 2000 years after creation. This date was chosen based on the assumption that these geographically widespread fossils represented post-Babel populations. Of the eight post-Flood patriarchs whose deaths are recorded in the Masoretic text of Genesis 11, all died between 1993 and 2184 years after creation. Thus, a date of death of 2000

years after creation for the fossils in question appears reasonable.

## Results

**Differences.** Based on the alignment of all mtDNA genomes in this study (excluding the artificial Eve consensus), the average single nucleotide difference (SND, transitions plus transversions) between Neandertal and *Homo sapiens* was 190.9 ( $\sigma = 5.8$ ). The average SND between Denisovans and *Homo sapiens* was 370.8 ( $\sigma = 4.6$ ), and the average SND between Denisovans and Neandertals was 355 ( $\sigma = 2.8$ ). Compared to the two chimpanzee mtDNA genomes, the Neandertals were slightly more similar (average SND = 1393.2,  $\sigma = 3.4$ ) than the Denisovans (average SND = 1419.5,  $\sigma = 4.1$ ) or *Homo sapiens* (average SND = 1418.2,  $\sigma = 7.1$ ) (Table 1).

DeWitt and Skinner (2001) argued that polymorphisms make Neandertals appear artificially more different from modern humans than they otherwise would. Fixed differences were therefore examined, defined as differences where all members of one group had the same nucleotide in one position, but all members of a second group had a different nucleotide at the same position.

Based on the alignment of the full dataset of mtDNA genomes (excluding the artificial Eve sequence), the six Neandertals were found to have 104 fixed SNDs when compared to *Homo sapiens* (Table 2). The two Denisovans had 265 fixed SNDs when compared to *Homo sapiens*, but 349 fixed SNDs when compared to the Neandertals. Compared to chimpanzees, there were 1189 fixed SNDs when compared to *Homo sapiens*, 1370 when compared to Neandertals, and 1423 when compared to Denisovans. These results appear to suggest that Denisovans are more similar to *Homo sapiens* than to Neandertals (265 vs. 349 fixed SNDs respectively) and that *Homo sapiens* are more similar to chimps than to Denisovans (1189 vs. 1423 fixed SNDs respectively), but this is simply an artifact of the large sample size of *Homo sapiens*, which allows detection of rare polymorphisms.

Since Criswell's (2009) concerns about ancient DNA especially highlighted spontaneous deamination of cytosine that results in the substitution of thymine for cytosine, fixed SNDs based only on transversion differences were also evaluated (Table 3). Between *Homo sapiens* and Neandertals, there were seven fixed transversion SNDs, and between *Homo sapiens* and Denisovans, there were 19 fixed transversion SNDs. In contrast, when compared to chimps, there were at least six times as many fixed transversion SNDs as there were between members of genus *Homo*. Denisovans had 118 fixed transversion SNDs when compared to chimps, and Neandertals had 120. There were 246 fixed transversion SNDs between chimps and *Homo sapiens*.

Looking at the distribution of all pairwise SNDs between two *Homo sapiens*, between *Homo sapiens* and Neandertals, and between *Homo sapiens* and Denisovans (Figure 1), there is no overlap between the *sapiens*-Neandertal SNDs and the *sapiens-sapiens* SNDs. The *sapiens*-Denisovan SNDs do not overlap the *sapiens*-Neandertal SNDs or the *sapiens-sapiens* SNDs. Looking only at transversion SNDs, there is a slight overlap between the distribution of *sapiens-sapiens* SNDs and the *sapiens*-Neandertal SNDs, although the two distributions are still distinct. The distribution of *sapiens*-Denisovan transversion SNDs do not

**Table 1.** Mean and standard deviations of single nucleotide differences for all mitochondrial genomes in this study

	<i>Homo sapiens</i>	Neandertals	Denisovans
<i>Pan</i>	1418.2 ( $\sigma = 7.1$ )	1393.2 ( $\sigma = 3.4$ )	1419.5 ( $\sigma = 4.1$ )
Denisovans	370.8 ( $\sigma = 4.6$ )	355.0 ( $\sigma = 2.8$ )	
Neandertals	190.9 ( $\sigma = 5.8$ )		

**Table 2.** Fixed single nucleotide differences for all mitochondrial genomes in this study

	<i>Homo sapiens</i>	Neandertals	Denisovans
<i>Pan</i>	1189	1370	1423
Denisovans	265	349	
Neandertals	104		

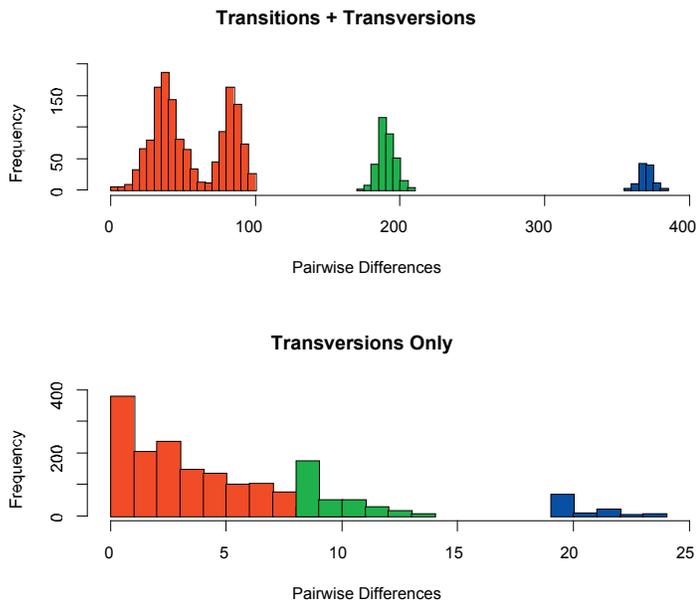
**Table 3.** Fixed transversion single nucleotide differences for all mitochondrial genomes in this study

	<i>Homo sapiens</i>	Neandertals	Denisovans
<i>Pan</i>	246	120	118
Denisovans	19	21	
Neandertals	7		

overlap either the *sapiens-sapiens* transversion SNDs or the *sapiens*-Neandertal transversion SNDs.

The ratio of transversions to transitions appears to be largely consistent in pairwise comparisons of *Homo sapiens* to other *Homo sapiens* (average ratio = 0.0647,  $\sigma = 0.039$ ), *Homo sapiens* to Neandertals (average ratio = 0.0545,  $\sigma = 0.0081$ ), and *Homo sapiens* to Denisovans (average ratio = 0.0585,  $\sigma = 0.0045$ ) (see Figure 2). When compared to animals, however, the average transversion/transition ratio was significantly higher for pairwise comparisons of *Homo sapiens* to chimpanzees (average ratio = 0.924,  $\sigma = 0.0011$ ) and *Homo sapiens* to gorilla (average ratio = 0.148,  $\sigma = 0.0013$ ).

**Substitution Rates.** To infer substitution rates and divergence dates, a phylogeny of the full set of 63 human (*Homo sapiens*, Neandertal, and Denisovan) mtDNA genomes was constructed using MEGA 5. The phylogeny used a Tamura-Nei (1993) substitution model, based on a model selection Bayesian Information Criterion of 65,963 (model selection results not shown). Rate variation was modeled using a discrete gamma

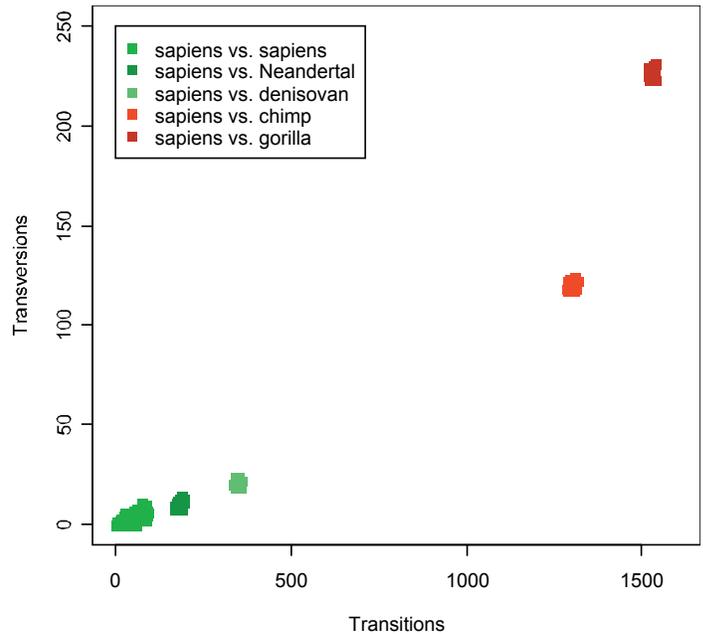


**Figure 1.** Pairwise single nucleotide differences (SNDs) measured from an alignment of complete mitochondrial genomes from 54 *Homo sapiens*, six Neandertals, and two Denisovans. Shown are the SNDs for *Homo sapiens*-*Homo sapiens* comparisons (red), *Homo sapiens*-Neandertal comparisons (green), and *Homo sapiens*-Denisovan comparisons (blue).

distribution with five categories and a shape parameter of 0.1. The tree with the highest log likelihood (-34143.7) is shown in Figure 3 with a midpoint rooting.

The simplest method of inferring divergence dates would be to calibrate the midpoint root at 6000 years ago (i.e., at Creation), keeping in mind that the result should not be entirely accurate given that the Neandertal, Denisovan, and Kostenki individuals are not from extant populations. The result of this calibration implies that most recent ancestor for modern humans lived approximately 1300 years ago and that the Neandertals branched from modern humans some 3000 years ago. These dates are clearly impossible, and thus the simple calibration model is invalid, as expected. Indeed, when the molecular clock was tested for these individuals using a log likelihood test, the hypothesis that the rates were equal throughout the tree was rejected (log likelihood with clock: -32187.0; log likelihood without clock: -32131.1;  $p < 7.8 \times 10^{-5}$ ).

A second phylogeny of eight approximately contemporaneous ancient mtDNA genomes was constructed to assess the molecular clock at a particular point in time (see Methods). The maximum likelihood phylogeny with the highest log likelihood (-25109.2) is shown with a midpoint rooting in Figure 4. The phylogeny was constructed using a Tamura-Nei (1993) substitution model (selected based on a Bayesian Information Criterion of 50,443), with a five category, discrete gamma distribution of rates among sites (shape parameter = 0.05). For this phylogeny, the null hypothesis that all taxa are evolving at roughly the same rate could not be rejected in a log likelihood test (log likelihood with clock:



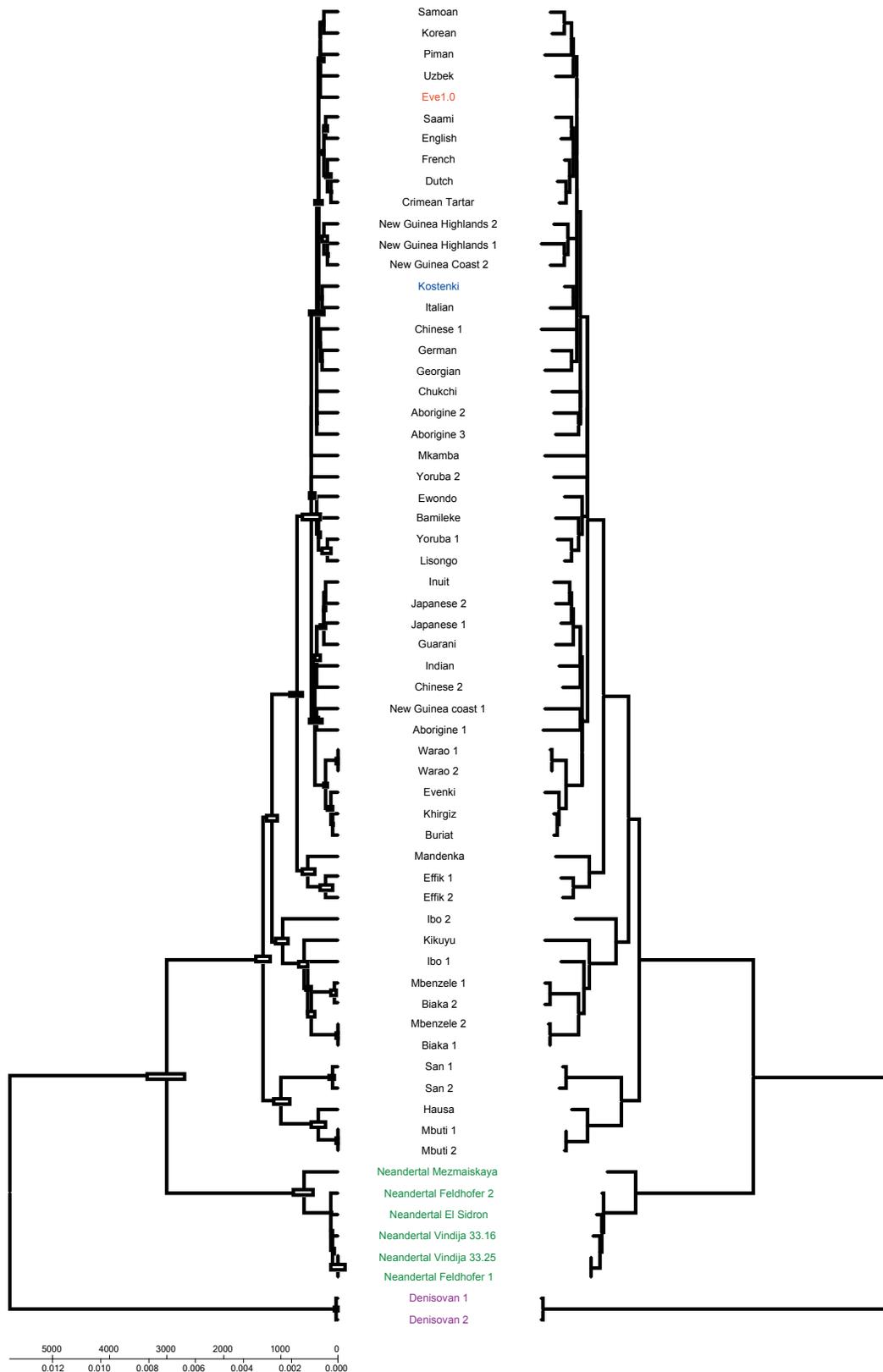
**Figure 2.** Transitions and transversions measured from an alignment of complete mitochondrial genomes from 54 *Homo sapiens*, six Neandertals, two Denisovans, two chimpanzees, and one gorilla.

-25111.8; log likelihood without clock: -25109.2;  $p < 0.54$ ). The phylogeny was therefore calibrated by placing the midpoint root at 2000 years before the deaths of the taxa (see Methods).

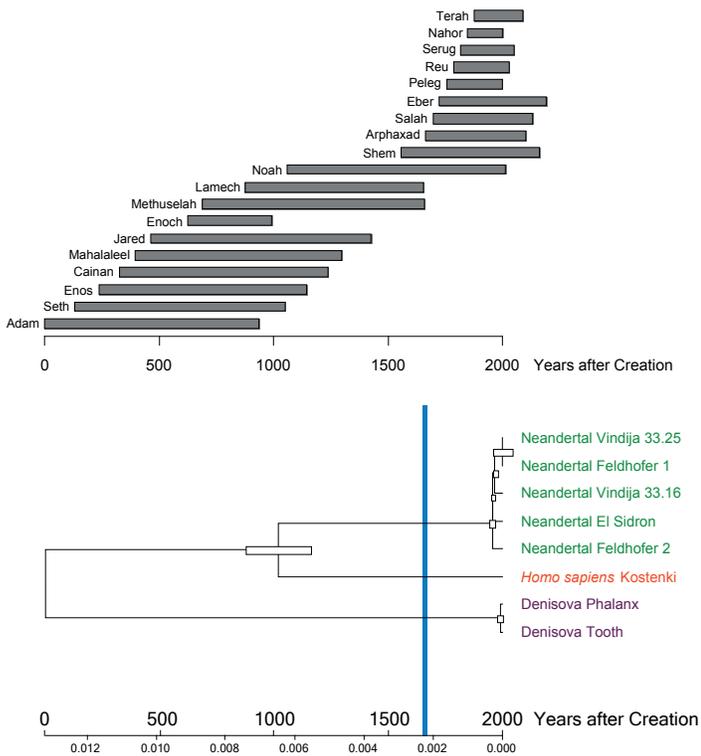
The calibrated phylogeny places the divergence of Neandertal and ancient *Homo sapiens* mtDNA before the Flood, about the time of the birth of Noah's father Lamech. If the individuals from which these ancient mtDNA sequences were taken died approximately 2000 years after Creation as assumed, then they could have been born at nearly any time after the Flood, since the post-Flood patriarchs listed in Genesis 11 died around 2000-2100 years after Creation (see methods). Thus, the number of generations separating them from Eve could be anywhere from eleven to nineteen. The average number of SNDs between Neandertals and the two Denisovan sequences is 376.4, which yields an average substitution rate of 9.9 substitutions/generation (assuming eleven generations divergence) or 17.1 substitutions/generation (assuming nineteen generations divergence). Both Denisovan mtDNA genomes differed from the Kostenki *Homo sapiens* mtDNA genome by 385 substitutions, which yields an average substitution rate of 10.1 substitutions/generation (assuming eleven generations divergence) or 17.5 substitutions/generation (assuming nineteen generations divergence).

## Discussion

Based on an exacting analysis of Neandertal hypervariable regions I and II, DeWitt and Skinner (2001) concluded that Neandertal mtDNA "cannot be excluded from the range of human



**Figure 3.** Maximum likelihood phylogeny inferred from an alignment of complete mitochondrial genomes from 54 *Homo sapiens* (black), six Neandertals (green), two Denisovans (purple), and the artificial consensus sequence Eve 1.0 (red). The position of the ancient Kostenki sequence is highlighted in blue. Phylogenies are shown with (right) and without (left) molecular clock calibration, assuming that the midpoint root is 6000 years before present.



**Figure 4.** Maximum likelihood phylogeny inferred from an alignment of complete mitochondrial genomes from five Neandertals (green), two Denisovans (purple), and one *Homo sapiens* (red), calibrated at the midpoint root to a divergence date of 2000 years before the deaths of the individual taxa. The date of the Flood according to the Masoretic chronology of Genesis 5 and 11 is shown in blue, and the lifespans of the Genesis 5 and 11 patriarchs are shown above the phylogeny.

variability.” The present results do not support that conclusion. Instead, 104 fixed SNDs were found between the complete mitochondrial genome sequences of six Neandertal specimens and 54 *Homo sapiens*. Even if cytosine degradation is a serious source of sequence error in ancient sequences, there are still seven fixed transversion SNDs between Neandertals and humans, none of which can be the result of cytosine degradation (which would result in transitions not transversions).

Furthermore, the distribution of pairwise SNDs also indicates that Neandertals are outside the range of *Homo sapiens* mtDNA variability. Total SNDs show no overlap between the pairwise SNDs of *Homo sapiens-Homo sapiens* comparisons and Neandertal-*Homo sapiens* comparisons. Even if many of these pairwise differences are caused by degradation (which is unlikely), the distribution of transversion SNDs between Neandertals and *Homo sapiens* is not within the range of intra-*Homo sapiens* variation.

The differences between Neandertal and *Homo sapiens* mtDNA are only amplified by comparison to Denisovan

mtDNA. Denisovan mtDNA had 265 total fixed SNDs and 19 fixed transversion SNDs when compared to *Homo sapiens*. The pairwise distribution of Denisovan-*Homo sapiens* SNDs was well outside the range of *Homo sapiens-Homo sapiens* SNDs, when considering either total SNDs or transversions only (Figure 1).

Despite these differences, there remains evidence that both Neandertals and Denisovans were human (i.e., descended from Adam and Eve). In the case of the Neandertals, recent hominid baraminology studies unequivocally place them in the human holobaramin (Wood 2010), and evidence of introgression (Green et al. 2010) supports this based on Marsh’s (1947) interbreeding criterion for identifying membership in a created kind. For Denisovans, evidence of the common humanity with *Homo sapiens* is limited to genetic evidence of introgression into modern Melanesian populations from Denisovans, but this again would be considered powerful evidence by creationists applying Marsh’s (1947) interbreeding criterion.

Indeed, the common humanity of Denisovans, Neandertals, and *Homo sapiens* would seem to be further supported by the striking differences between these three human groups and non-human apes (Figure 2). Whereas Robinson (1997) noted a very narrow distinction between turtles and non-turtles in his analysis of turtle mtDNA, there is a vast gap between humans and gorillas and between humans and chimpanzees (Figure 4). These differences extend even to the ratio of transversions to transitions, which is substantially higher in comparisons of *Homo sapiens* to non-human apes than in comparisons of *Homo sapiens* to Denisovans and Neandertals. Whether these differences should be considered evidence of discontinuity should be addressed in future studies.

Previously, creationists have emphasized the work of Parsons et al. (1997) as evidence that mtDNA diversity in extant *Homo sapiens* really could be traced back to an ancestor living around 6000 years ago (Wieland 1998; Kulikovskiy 2000; DeWitt 2003). Since these ancestral estimates were based entirely on the similarity of *Homo sapiens* mtDNA, we could have anticipated difficulties from incorporating more divergent lineages (or even other human species) into the human mtDNA tree. Indeed, the present results indicate that the divergence rate estimated by analysis of full mtDNA genomes is 333 times faster than the rate of 1 substitution in 33 generations estimated by Parsons et al. (1997). If all human mtDNA diverged at 1 substitution in 33 generations, then the divergence of *Homo sapiens* and Denisovans would have required 6352.5 generations, based on the average difference of 385 SNDs between the mtDNA genomes of the Kostenki *Homo sapiens* and the Denisovans. At an average generation time of at least 20 years, that translates to a date of roughly 127,050 years ago. Consequently, inclusion of the Denisovans and Neandertals in the human family tree requires much higher substitution rates than even Parsons et al. (1997) estimated.

That is not to say that the present substitution rate estimates are without error. The rate estimates made here are sensitive to the assumptions of 11-19 generations of divergence and the midpoint root. For example, if the ancient individuals whose mtDNA has been sequenced died some time after 2000 years after Creation, the estimated substitution rates would be reduced. Alternatively, if *Homo sapiens* and the Denisovans share a common ancestor more recent than the original creation, the estimated substitution rates would go up. However, any changes based on changing the

divergence time (measured in years or generations) would not appreciably alter the substitution rates, certainly not to the point of being consistent with Parsons et al.'s estimate of 1 substitution in 33 generations.

The more significant assumption used to estimate substitution rates here was the midpoint root. The midpoint root strongly contrasts with Carter et al. (2008), who argued that a consensus of modern *Homo sapiens* called Eve 1.0 is "nearly identical to the real Eve mitochondrial sequence." Conclusively testing the Eve 1.0 sequence (or any other sequence) as the ancestral sequence for the human holobaramin is impossible for theoretical reasons. Given a selection of modern sequences, there is simply no way to know conclusively what the true ancestral sequence was. However, the Eve 1.0 sequence is unusual in that it is a member of the Eurasian R haplogroup. Thus, for Eve 1.0 to be the ancestor of all humans, substitution rates must be accelerated in African haplogroups but even more for the Neandertal and Denisovan lineages. Studies of African mtDNA have shown that haplogroups L2a and L2c seem to be evolving more slowly, which would not be consistent with the ancestral status of Eve 1.0 (Howell et al. 2004).

In this analysis, the maximum likelihood phylogeny places the Eve 1.0 sequence on a clade of Eurasian *Homo sapiens* sequences that includes the Kostenki sequence (Figure 3). In contrast, the midpoint rooting separates the human sequences into two clades, Denisovan and non-Denisovan, with the deepest root for the modern *Homo sapiens* sequences separating a clade of African sequences (Haplotype L) from a much larger clade of African and non-African sequences. If Eve 1.0 is to be considered the true root of the human tree, there would have to be extraordinary rate variation across most of the human mitochondrial tree, such that some modern and ancient sequences have changed very little, while other branches have changed dramatically. The Denisovan and Neandertal mtDNA genomes in particular must have resulted from extraordinarily high substitution rates compared to other branches of the human mtDNA tree.

The extraordinary rate variation required by rooting the tree with Eve 1.0 is not supported by the molecular clock test on the ancient mtDNA genomes (Figure 4). In that test, the Neandertal fossils sampled from across the European range of Neandertals all yielded mtDNA sequences that did not differ substantially. Thus, if Eve 1.0 really were the root of the human tree, we would have to assume that the Neandertals diverged extremely rapidly but did so in a coordinated way that mimicked a molecular clock. Such an assumption stretches credibility. Consequently, it would seem that Eve 1.0 is just a consensus of modern *Homo sapiens* mtDNA genomes and not a true ancestral sequence.

The present research does however support non-constancy of the human mtDNA molecular clock. Specifically, the presence of the Kostenki sequence so soon after the Flood suggests that much of the human mtDNA diversity (such as the African L haplotypes) was already present at the time of the Kostenki individual died. The similarity of the Kostenki mtDNA genome to extant Eurasian genomes suggests that there has been little change since the Kostenki individual died soon after the Flood. Thus, an accelerated substitution rate leading up to the Kostenki individual is necessary to accommodate the Neandertals and Denisovans in the human mtDNA tree, but the subsequent substitution rate must have returned to something more similar to the empirical

estimate of Parsons et al. (2007) to prevent any substantial divergence building up in the remaining human population in the approximately 4000 years after the Kostenki individual's death.

The presence of substantial human mtDNA diversity at 2000 years after Creation also suggests that some modern *Homo sapiens* mtDNA types might pre-date the Flood. Assuming that all the women on the Ark were homoplasmic, only four human mtDNA lineages could have survived the Flood (Wood 2008). The presence of the Denisovan, Neandertal, and *Homo sapiens* lineages soon after the Flood is therefore consistent with pre-Flood divergence, since each mtDNA type could have survived in a separate woman aboard the Ark. If, however, multiple *Homo sapiens* mtDNA types pre-date the Flood, we must either assume that at least one of the women aboard the Ark was heteroplasmic or that the divergence of the Denisovans and *Homo sapiens* occurred after Creation. The former scenario would require and even greater acceleration to the human mtDNA molecular clock.

It is tempting to speculate that the occurrence of a period of accelerated nucleotide substitutions around the time of the Flood is somehow linked to the rapid intrabaraminic diversification that is suggested to be taking place at the same time (Wood and Murray 2003, chap. 11). Previous theories for explaining rapid diversification emphasized chromosomal rearrangement (Wood 2003) or non-random mutations (Lightner 2009). If the results in this present study are correct, then there may have also been a period of accelerated mutation and substitution around the time of the Flood. Whether that period was one of the causes of intrabaraminic diversification remains to be demonstrated.

Finally, it should be emphasized that the present results apply only to the divergence of mitochondrial DNA, not to other parts of the genome nor to the human lineages themselves. As noted previously (Wood 2008), different parts of human genomes appear to have different genetic histories. Note especially in this regard the conflicting phylogeny between the Denisovan nuclear genome, which supports a sister taxon relationship with the Neandertals (Reich et al. 2010), and the Denisovan mitochondrial genome, which supports a sister taxon relationship between *Homo sapiens* and Neandertals (Krause et al. 2010b). Although the present study is limited to mitochondrial DNA, it is likely that the accelerated substitution rates proposed here will be supported in studies of nuclear genes and DNA from other holobaramins altogether, if a period of rapid nucleotide mutation really occurred.

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