International Journal of Biology; Vol. 11, No. 1; 2019
ISSN 1916-9671 E-ISSN 1916-968X
Published by Canadian Center of Science and Education

A Possible Glimpse at the Role of Naturally-Occurring Radiation as a Contributing Factor to Genetic Variance among Populations of Living Organisms

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Received: November 3, 2018 Accepted: December 4, 2018 Online Published: December 17, 2018

doi:10.5539/ijb.v11n1p42 URL: https://doi.org/10.5539/ijb.v11n1p42

Abstract

A fundamental question in evolutionary biology is understanding why some groups of organisms are highly diverse. Species diversity is the result of the balance between speciation and extinction whereas morphological disparity is primarily a consequence of adaptation. Radioresitance, among other adaptive features found in organisms, can be genetically determined and inherited. This study explores the possible consequences that naturally-occurring radiation might impose on the variance and composition of population genetics. To evaluate these possibilities, a simple statistical experiment is conducted involving 49 cytochrome gene sequences belonging to two distinct populations of eukaryotic organisms that reside on opposite ends of the radioresistance spectrum. The results show a distinct correlation between higher and lower degrees of genetic variance among these two groups, where the order, group or population that has shown increased hypersensitivity to radiation also has an increased rate of molecular evolution due to a higher mutation rate.

Keywords: Radioresistance, Radiation-Induced Evolution, Population Genetics, Comparative Sequence Analysis, Computational Phylogenetics

1. Introduction

While the effects of radiation on the cell biology of individual organisms have been widely investigated, there has been a dearth of publications involving the role of naturally-occurring radiation in the evolutionary development of living populations. For one, the sun emits short-wave UV radiation that ionizes at about 120 nm to 10 nm (Hori et al., 2014), and it can disrupt the normal chemical processes of cells, causing them to become damaged, to grow abnormally or to die. Moreover, radiation can create mutations in DNA that could be inherited over successive generations (Adewoye et al., 2015; Verhofstad et al., 2008; Barber et al., 2002; Tatebe et al., 2017; Linschooten et al., 2013; National Research Council, 2006). Two nucleotide bases in DNA-cytosine and thymine-are most vulnerable to the effects of radiation (Pray, 2008). For low levels of radiation exposure (or non-ionizing radiation), the biological effects are so small they are thought to cause little or no damage to cells (Upton et al., 1992).

Over the course of Earth's history, changes to the atmosphere must have played a key role in the conditions under which life formed and evolved (Cnossen et al., 2007). It is estimated that contemporary organisms would be killed in a matter of seconds if exposed to the full intensity of solar radiation from a primitive Earth (Madronich et al., 1998; Zepp et al., 2007; Hessen, 2008; Samson, 1977). Today, of course, living organisms are protected by an atmospheric ozone layer that effectively absorbs light at short wavelengths (Samson, 1977). Despite the presence of a modern atmosphere, various known groups of eukaryotic life forms, mostly those belonging to the invertebrate phylum, can tolerate considerable levels of acute ionizing radiation (Fuller et al., 2015; Adam-Guillermin et al., 2018). Other phyla, primarily mammals, are much more hypersensitive in that respect (Douglass & Jeffery, 2014). The range of radioresistance in eukaryotic organisms is considerable and, in some cases, quantifiable. This stark contrast might lead us to propose the following inquiry: What role, if any, does naturally-occurring radiation (atmospheric, background or terrestrial) play in shaping the outcome of living systems over the extent of evolutionary time? For purposes of this investigation, two distinct orders of eukaryotic organisms that reside on opposite ends of the radioresistance spectrum are examined: scorpions and rodents.

Scorpions are largely resistant to the effects of atmospheric radiation. For instance, their exoskeleton makes it effective at reflecting UV radiation (Husemann et al., 2012); better shielding its organs from radiation damage and, subsequently, protecting its cells from genetic mutation. Scorpiones occur in a vast array of different environments that put it in proximity to the most severe extremes of atmospheric radiation on Earth (Gaffin & Barker, 2014). A small number of Scorpiones species have even been documented to resist doses of radiation as high as 154,000 roentgens (D'Suze, 1990; Armas, 2001). What's more intriguing in this discussion, scorpion morphology has changed relatively little since their first appearance in the fossil record (Pryke, 2016).

Unlike scorpions, rodents are moderate to highly susceptible to the effects of naturally-occurring radiation. Ongoing experimentation has shown that rodents have a median lethal dose (LD_{50}) of 7.5 against ionizing radiation exposure (Srinivas et al., 2015), whereas other mammalians range between 3.5 and 12 (Srinivas et al., 2015). During the Eocene, rodents evolved and diversified into a wide range of subfamilies that make up about 40% of all mammalian species and they too occur on almost every continent (Churakov et al., 2010). Not only is their proliferation extensive but also the rates of DNA evolution vary significantly among lineages, which has hindered attempts to reconstruct a robust phylogeny (Churakov et al., 2010). Most importantly, several studies have largely shown that an increased rate of molecular evolution in rodents is found to be entirely the consequence of a higher mutation rate as compared to other species, families, and genera (Weinreich, 2001; Schlager et al., 1971; Uchimura et al., 2015; Drake et al., 1998; Lindsay et al., 2016).

Genetic mutation is the raw material needed for evolution to occur. Natural selection acts upon genetic variation to create the diversity around us. Whether by direct exposure to or by indirect selective pressures from, this study explores the possible consequences that naturally-occurring radiation might impose on the variance and composition of population genetics. Furthermore, it is proposed that naturally-occurring radiation may be implicated in creating genetic variance among different populations of living organisms over extended evolutionary time. To evaluate strength of this hypothesis, a simple statistical experiment is conducted involving 49 cytochrome gene sequences examined in phylogenetic context, pairwise dissimilarity ratios, and molecular clock estimates.

2. Material and Methods

2.1 Sequence Selection

To begin this experiment, 31 cytochrome b sequences were selected representing a small portion of rodent species that occur or originate within hemispherical proximity; whereas the 18 scorpion cytochrome COI sequences are more geographically widespread and diverse, as to include both hemispheres and six continents. (Note 1) The two most commonly used genetic loci in species identification are the cytochrome oxidase I gene (COI) and the cytochrome b gene (Tobe et al., 2010). In terms of phylogenetic analysis, the application of cytochrome sequences is often selected for reasons involving sequence variability and the availability of related sequences from publicly curated databases (Tobe et al., 2010). Both cytochrome gene types utilized in this investigation are also practical due to their highly detectable rates of mutation. This makes it a good candidate for pairwise comparisons and phylogenetic reconstruction among closely related species.

Each raw sequence was obtained from the NCBI nucleotide databank. BLAST similarity searches were conducted to obtain homologous sequence candidates among closely related groups. (Note 2) FASTA format was preferable in this context, as it improves the ability to use a wider range of scoring matrices and facilities merging annotations into alignments (Pearson, 2013). Lastly, two distinct FASTA files containing a total of 49 raw sequences were compiled and later used toward sequence alignment and phylogenetic analysis.

2.2 Sequence Alignment & Phylogenetic Reconstruction

As it corresponds to the following procedures, Kalign was utilized for multiple sequence alignment (MSA) and pairwise comparisons; while PHYLIP neighbor-joining method, a distance matrix algorithm, was employed toward phylogenetic reconstruction. (Note 2) An accurate and fast MSA algorithm, Kalign is a dependable algorithmic selection for purposes of obtaining highly-robust alignments (Lassmann & Sonnhammer, 2005). Kalign is an extension of Wu-Manber approximate pattern-matching algorithm, based on Levenshtein distances. This strategy enables Kalign to estimate sequence distances faster and more accurately than other popular iterative methods. Lassmann and Sonnhammer (2005) show that Kalign is about 10 times faster than ClustalW and, depending on the alignment size, up to 50 times faster than other iterative methods; Kalign also delivers better overall resolution (Lassmann & Sonnhammer, 2005).

PHYLIP neighbor-joining method can be used to generate highly probable diagrams amid scenarios involving low degrees of variance, regardless of alignment size. Selected for these tree-building exercises, PHYLIP

neighbor-joining is an accurate and statically consistent polynomial-time algorithm that does not assume that all lineages evolve at the same rate, and it constructs a tree by successive clustering of lineages, setting branch lengths as the lineages join [where a set of n taxa requires n-3 iterations; each step is repeated by (n-1) x (n-1)] (Felsenstein, 1981; Purvis, 1995). This method incorporated a set of default parameters for distance matrix model F84. Additional bootstrapping compilers were applied to a ratio of 70%-75% for better overall resolution. For reference purposes, the following formula demonstrates a standard neighbor-joining Q-matrix algorithm:

$$Q(i,j) = (n - 2)\ d(i,j) - \sum \{n,\ k = 1\}\ d(i,k) - \sum \{n,\ k = 1\}\ d(j,k) \quad (1)$$

Pair to node (distances):

$$(f,u) = \tfrac{1}{2}\ d(f,g) + \tfrac{1}{2}(n - 2)\ [\ \sum \{n,\ k = 1\}\ d(f,k) - \sum \{n,\ k = 1\}\ d(g,k)\ ] \quad (2)$$

Taxa to node (distances):

$$d(u,k) = \tfrac{1}{2}\ [\ d(f,k) + d(g,k) - d(f,g)\ ] \quad (3)$$

3. Results

3.1 Pairwise Dissimilarity Ratios

After each sequence alignment was consolidated, a series of pairwise dissimilarity datasets were calculated using the total length of each cytochrome sequence between any two organisms and divided by the length of the total genomes of the organism in row. The total ratios are then used as a basis for the intensity of genomic discrepancy (or variance) among organisms of the same order. Specifically, for each measurement taken, percentages of pairwise dissimilarity were computed, a broader median within homologous sequences determined, and those figures were then compared against each respective group, lineage or order.

Table 1. Pairwise comparison of 31 cytochrome b sequences representing order Rodentia (in no particular order)

Annotation Number / Species / Base Pair	Consensus Percentage
>AB109397.1 Apodemus draco (1140 bp)	1.00
>KX790791.1 Mus cookii (1144 bp)	0.85
>AF159396.1 Mus poschiavinus (1144 bp)	0.85
>FR751074.1 Mus cypriacus (1140 bp)	0.85
>AB819920.1 Mus musculus (1140 bp)	0.85
>AF520637.1 Mus caroli (1191 bp)	0.81
>AY057808.1 Mus macedonicus (1145 bp)	0.85
>AF159397.1 Mus spicilegus (1144 bp)	0.85
>AB033700.1 Mus spretus (1140 bp)	0.86
>AJ698872.1 Mus famulus (1140 bp)	0.85
>AB125779.1 Mus fragilicauda (1140 bp)	0.87
>AB125777.1 Mus terricolor (1140 bp)	0.86
>KY587424.1 Mus booduga (1143 bp)	0.86
>AJ233955.1 Acomys minous (1141 bp)	0.80
>AJ233957.1 Acomys cilicicus (1141 bp)	0.80
>Z96053.1 Acomys cahirinus (1141 bp)	0.80
>EF187792.1 Acomys wilsoni (1141 bp)	0.80
>EU349734.1 Apodemus semotus (1136 bp)	0.91
>FR775869.1 Rattus andamanensis (1143 bp)	0.85
>JQ823422.1 Rattus rattus (1143 bp)	0.84
>DQ191487.1 Rattus praetor (1137 bp)	0.83
>JQ793903.1 Rattus tanezumi sladeni (1137 bp)	0.84
>KJ592784.1 Bandicota savilei (1139 bp)	0.82
>U87525.1 Heterocephalus glaber (1122 bp)	0.75
>KY753976.1 Diplothrix legata (1125 bp)	0.83
>KJ592782.1 Bandicota indica (1139 bp)	0.82
>KY587421.1 Bandicota bengalensis (1140 bp)	0.82
>KC735127.1 Rattus norvegicus (1143 bp)	0.83
>Z96068.1 Acomys spinosissimus (1141 bp)	0.80
>JN247708.1 Acomys muzei (1140 bp)	0.81

Results obtained from each dataset are shown in Table 1 and Table 2. Of the global measurements taken, the order Rodentia contained the highest total dissimilarity ratio, falling within the range of 9% to 25%; and a median score of 84%. Meanwhile, order Scorpiones ranged between 1% and 9% with a median score of 98%. Measurements of Rodentia also show far wider concentrations of diffused similarity nearest the middle of a bell curve and disperses in opposite directions relative to the mean. Rodentia as a group produced more total variance and those results are visually represented on a dot plot distribution shown on Figure 1. Each raw set (Rodentia, Scorpiones) generated a mean of 84, 98 and standard deviation of 4.2648, 2.4162 (see Figure 2 & Figure 3) (Note 3).

Table 2. Pairwise comparison of 18 cytochrome COI sequences representing order Scorpiones (in no particular order)

Annotation Number / Species / Base Pair	Consensus Percentage
>AY156582.1 Pandinus imperator (658 bp)	1.00
>AY156575.1 Heterometrus swammerdami (658 bp)	0.99
>AY156572.1 Heterometrus fulvipes (658 bp)	0.99
>AY156574.1 Heterometrus spinifer (658 bp)	0.99
>AY156573.1 Heterometrus laoticus (658 bp)	0.98
>AY156578.1 Opistophthalmus carinatus (658 bp)	0.98
>AY156577.1 Opistophthalmus capensis (658 bp)	0.99
>AY156576.1 Opistophthalmus boehmi (658 bp)	0.99
>AY156579.1 Opistophthalmus holmi cytochrome (658 bp)	0.98
>KT188295.1 Scorpio fuscus (658 bp)	0.98
>KT188319.1 Scorpio kruglovi (658 bp)	0.97
>KT188221.1 Scorpio propinquus (658 bp)	0.97
>KT188328.1 Scorpio palmatus (658 bp)	0.98
>JQ514257.1 Hadogenes paucidens (653 bp)	0.93
>JQ514256.1 Hadrurus arizonensis (659 bp)	0.91

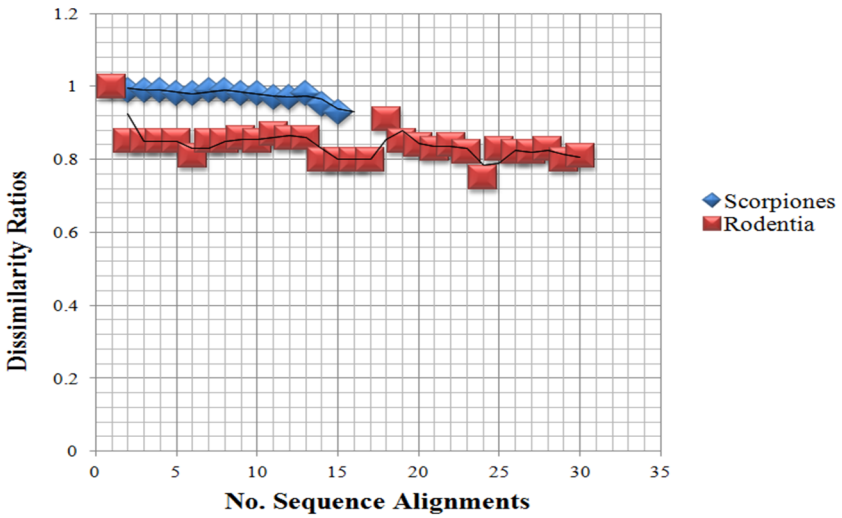


Figure 1. Dot plot distribution of pairwise comparisons between order Scorpiones and order Rodentia

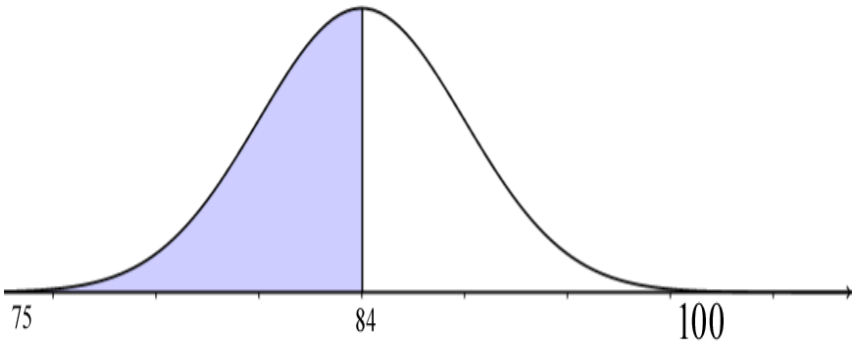


Figure 2. Bell curve distribution (Rodentia). Mean: 84, Standard Deviation: 4.2648, Normal Distribution: Pr(X<100) = 0.9819

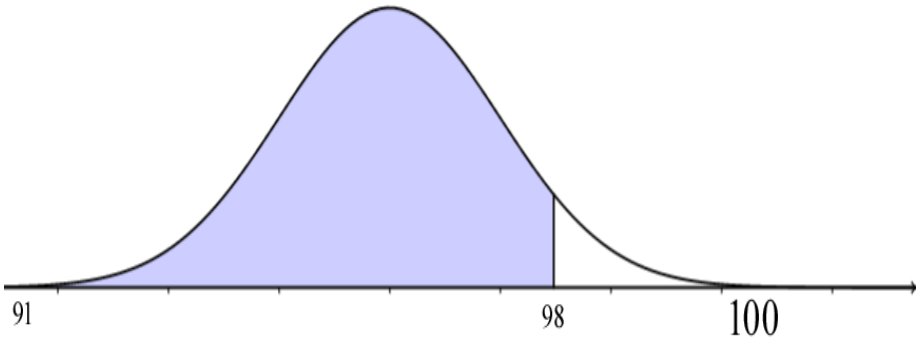


Figure 3. Bell curve distribution (Scorpiones). Mean: 98, Standard Deviation: 2.4162, Normal Distribution: Pr(X<100) = 0.7961

3.2 Implications of Evolutionary Trees

Phylogenetic comparative methods account for evolutionary history and explicitly model trait change along the branches of evolutionary trees (Dunn et al., 2018). The value of these methods relative to pairwise comparisons has been repeatedly shown in analysis of distance estimations between all possible pairs of sequences in a given dataset. Ultimately, the goal of improving distance estimation is to increase the accuracy of the reconstructed tree topology (Ninio et al., 2007). It then follows that each node estimate on the branch of a tree can represent a degree of confidence about the accuracy of any clade distribution within a given tree. In terms of reliability, bootstrapping is a common method for assessing confidence in the results of phylogenetic analysis. As other sources have noted, bootstrap proportions of ≥70% usually correspond to a probability of ≥95% that the corresponding clade is accurate (Yang, 2006; Hillis & Bull, 1993); and thus, the probability of inferring an accurate clade distribution increase.

The results are simulated under a null hypothesis model. The cladograms (or trees) derived from the original datasets do not show an excess of larger contrasts for duplication events, nor does it reject the null hypothesis. As shown on Figure 4 and Figure 5, the hierarchical placement of each clade, from outgroup within, are consistent with biogeographical distribution of contemporary species relative to each genus and each order (Jenkins, 2011; Monadjem et al., 2015; Lourenco, 2001). Moreover, the results agree with recent molecular studies that yielded significant evidence in support of Rodentia phylogeny (Blanga-Kanfi et al., 2009), with genus Apodemus and genus Mus appearing in the first branches on opposite ends of the outgroup; whilst genus Rattus and genus Heterocephalus are located among the intergroup clades (Figure 4). Studies done on Scorpiones phylogeny are more variable in terms of their agreement on clade placements (Sharma et al., 2018; Prendini & Wheeler, 2005). Here, both datasets are supported by strong comprehensive bootstrap values (approximately ≥94%) via PHYLIP neighbor-joining method for clade distributions.

3.3 Molecular Clock Estimates

The branch length estimates on Figure 5 suggest deep rooted ancestry whereas Figure 4 illustrates several short length clusters that could represent a series of rapid diversification events. This interpretation is based on the units of substitutions per site corresponding to each node on the diagram marked by their descending branch lengths in each lineage. Measures of lineage divergence are not a priori design to be best estimates of time, however, they are often good proxy for time and often interpreted as being measurements of time (Takezaki et al., 1995). To transform these lengths into a time scale, further information is required.

To start, rather than using a relaxed clock model that describes how each branch length l=r x t can be decomposed into a rate r and a time t (Lartillot et al., 2016), evolutionary time scales were postulated by utilizing the distances between neutral substitutions. The neutral substitution rate is often the best estimate we have of an underlying mutation rate (Lanfear et al., 2014). And, deciphering the average mutation rate in any given species should provide further opportunities for estimating species and population divergence times (Kumar & Subramanian, 2002).

Several studies have established the average mammalian genome mutation rate at ±2.2 × 10−9 per base pair per year (Kumar & Subramanian, 2002; Hodgkinson et al., 2011; Ellegren et al., 2003). Others have examined the null hypotheses of neutral mutation rates among Arachnida lineages and have come to some conflicting conclusions (Allio et al., 2017). The most commonly accepted mutation rate found in Scorpiones is 0.39 ± 0.94 per site × 109 years (Gantenbein & Eightley, 2004). By combining these external estimates to the results obtained from multiple sequence alignment, phylogenetic analysis, and pairwise comparisons, where Rodentia attained several clusters of short-length divergent times and higher rates of neutral substitutions, there is good support for the original inquiry. Patterns of diversification are more frequent among Rodentia lineages compared to the Scorpiones order (see Figure 4 & Figure 5).

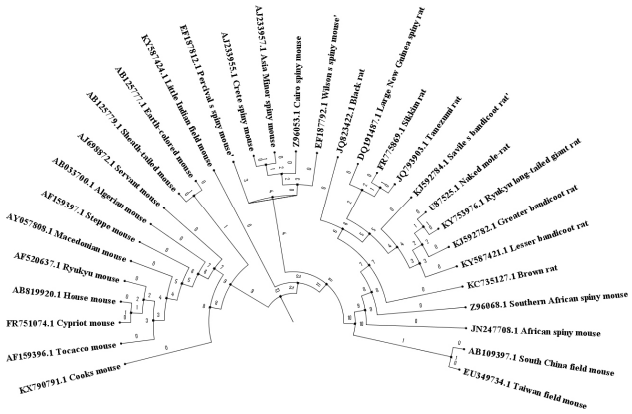


Figure 4. Phylogenetic reconstruction of 31 cytochrome b sequences representing order Rodentia

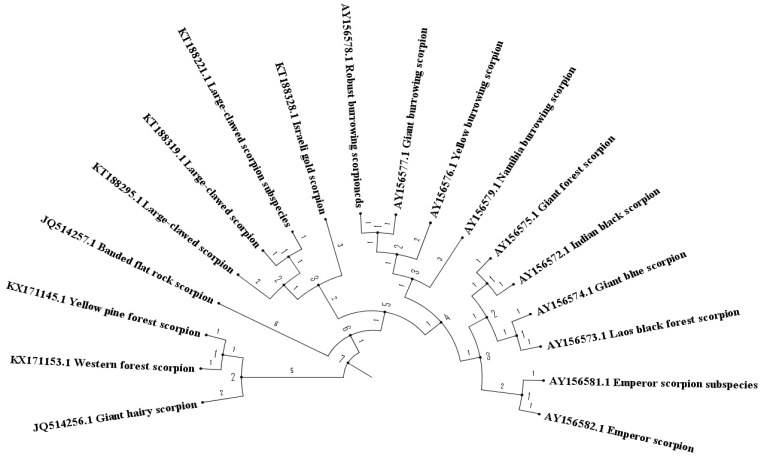


Figure 5. Phylogenetic reconstruction of 18 cytochrome COI sequences representing order Scorpiones

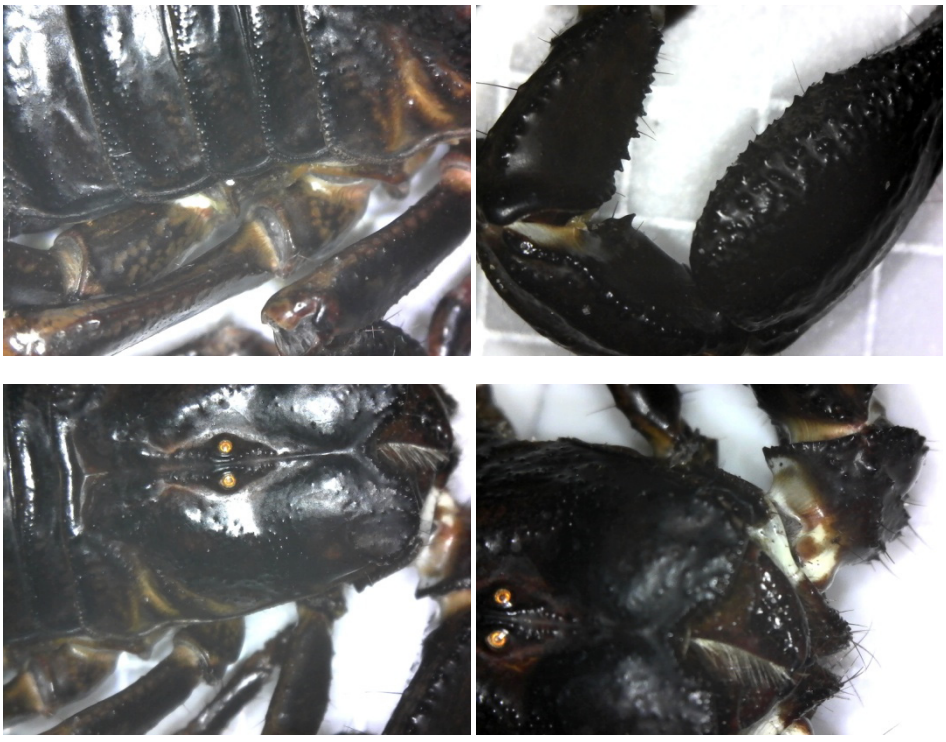
4. Discussions

A fundamental question in evolutionary biology is understanding why some groups of organisms are highly diverse. Species diversity is the result of the balance between speciation and extinction whereas morphological disparity is primarily a consequence of adaptation (Fabre et al., 2012). At the most fundamental level, mutations are essential to evolution. Every genetic feature in every organism was, initially, the result of a mutation (Carlin, 2011). All mutations that affect the fitness of future generations are agents of evolution (Carlin, 2011). Radioresitance, among other adaptive features found in organisms, can be genetically determined and inherited (Nöthel, 1987; Cordeiro et al., 1973; Ling & Endlich, 1989; Joiner, 1994).

Radioresistance is surprisingly high in many organisms. For example, the study of environment, animals and plants around the Chernobyl accident area has revealed an unexpected survival of many species, despite the high radiation levels (Deryabina et al., 2015). A Brazilian study in a hill in the state of Minas Gerais which has high natural radiation levels from uranium deposits, has also shown many radioresistant insects, worms and plants (Cordeiro et al., 2973). These rare examples give us a glimpse into radioresistance at a localized generational scale but say nothing about the possible influx of selective pressures that it imposes over long stretches of geological time.

We know organisms that are susceptible to radiation are often more inclined to its effects; such effects include those that cause permanent alterations of nucleotide sequences of the genome (or a genetic mutation, simply speaking). To this end, organisms that better resist the effects of radiation also have a relative fitness advantage to its damaging effects and it too may have an impact at the population level. Selective traits are inherited. Let's take another look at Scorpiones, for instance.

The scorpion's carboline-composed exoskeleton is thick and durable, providing good protection from a range of environmental threats (see Images 1-4). Carboline chemicals are found in many animal cells, not just scorpions, and are thought to be sunscreens that protect epidermal cells by reflecting or scattering UV radiation (Husemann et al., 2012). Research suggests that oxygen levels were much lower when the first ancestral scorpion lineages appeared during the Silurian period; a time when more UV radiation was able to reach the Earth's surface (Hsia et al., 2013). This might explain the evolution of carboline in the exoskeletons of modern day Scorpiones. In addition to its carboline-composed exoskeleton, other studies have demonstrated that internal antioxidant chemistry may be responsible for shielding its cells from radiation damage (Levin-Zaidman et al., 2003). What's more, scorpion morphology has changed relatively little since their first appearance in the fossil record (Pryke, 2016).



Images 1-4. Digital microscopic images of Scorpiones carboline exoskeleton at 30X. Species shown: *Heterometrus spinifer*

Longwave UV light is reflected as visible light in the green range. For this reason, scorpions appear to glow in darkness. Under direct sunlight, the fluorescence may impart a greenish tint to the scorpion's color (Smithsonian Institution, 2018). Studies have shown this feature to derive from certain molecules in one layer of the cuticle, the tough but somewhat flexible part of a scorpion's exoskeleton, which absorbs the longer wavelengths of UV light and emit it in different wavelengths that are visible at night as a blue-green glow (Ray, 2017).

An experiment led by Kloock et al. (2010) found that the fluorescence seemed to help scorpions detect and avoid UV light (Ray, 2017). The activity level of the test subjects in this study was measured by the number of transitions from the exposed to shadowed regions, and choice was measured by the percentage of time spent in the shadowed portion of the arena under IR-UV (Kloock et al., 2010). Kloock et al. interpreted their observations as evidence that fluorescence aids in the detection of and response to UV light.

Now, back to the topic of DNA. The evidence from cell science is further compelling. Consider the following: Aside from errors in cellular replication and oxidative damage (Crow, 2000), germline mutations can also occur due to exogenous factors that include chemical substances and ionizing radiation (Rahbari et al., 2016; Cai et al., 1995). One well-documented study describes how single exposure or multiple exposures to a low dose radiation induced a significant cytogenetic adaptive response in mouse germ cells (Cai et al., 1995). As noted above, Scorpiones are unique in that they combine traits for UV light avoidance along with a UV reflective exoskeleton and an internal metabolism fit for shielding radiation damage to cells. The hypersensitivity of the rodent anatomy, as with other members of the mammalian phylum, predisposes it to more of the DNA-damaging effects of naturally-occurring radiation.

Biogeography is another considerable element to this study worth mentioning. One of the objectives was to demonstrate a quantifiable degree of genetic variance between two distinct groups of organisms in conjunction with, or despite of, their geographical distribution. Furthermore, it might be expected that higher degrees of variance would be present among groups of closely related organisms that occur across wider ranges in a geographical context; and the reciprocal scenario could be assumed in like manner.

Both rodents and scorpions overlap in terms of their natural environments, exposing each form to nearly equal amounts of atmospheric radiation brought on from UV light. Surprisingly, despite a much lower ratio of total pairwise dissimilarity, members of group Scorpiones also happen to be more biogeographically extant. Moreover, it has also been shown that it's counterparts, namely those members belonging to order Rodentia, have an increased rate of molecular evolution due to a higher mutation rate. Rodentia is the most diverse order of placental mammals, with extant rodent species representing about half of all placental diversity (Blanga-Kanfi et al., 2009).

Thus, the question now becomes whether localized radiation events contribute to an increased rate of molecular evolution in organisms that lack radioresistance. And if so, what can we infer about the opposite case scenario? Studies like these pose several unique challenges. First, the data sampling used in this study is both incomplete and relatively small to draw any significant inferences about an entire phylum of species. And, in fact, much of this interpretation could be explained in the historical context of things like genetic drift, retrovirus insertions, or even the vacancies left by open ecological niches; just to name a few examples. While many of these mechanisms do play invaluable roles in species diversification, this study solely ventures to assess any possible association that may exist between naturally-occurring radiation and its implications on population genetics among groups of organisms that reside on far ends of the radioresistance spectrum.

To what extent to which population genetics becomes affected by natural-occurring radiation will require further investigation. Similar studies in laboratory settings have focused on microorganisms but rarely has it included multi-cellular eukaryotes. Indeed, an argument could even be raised about the timescale limitations involved with testing multi-cellular eukaryotes, coupled with the rarity of inheritable mutation, which would make such an experiment impractical in a controlled environment. Yet, there may be a reliable framework by which to detect, assess, and quantify radiation-induced evolution over successive generations. Before this paper concludes, let's elaborate further on a potential experiment design.

Imagine for moment, a more ideal experiment involving three homologous sets of fast-breeding model organisms that have adjusted their sexual reproductive patterns to satisfy remarkably short life cycles. Over the course of this experiment, each group is exposed to different levels UV radiation (perhaps ranging between 180 and 400 nm); except for the control group, which remains unexposed. Such an experiment could incorporate test subjects like Drosophila melanogaster or Acheta domestica, as they are known to be good candidates for studying a wide array of biological processes. After careful match-breeding over different generations, gene sequencing is applied to each line of offspring within the same test group or population. Complete sequences of

mitochondrial DNA could be a useful biomarker in this experiment, due to its tendency toward rapid mutation rates. Techniques in comparative sequence analysis can then reveal even the slightest biological change over a multitude of successive generations. In some ways and on a more simplified scale, this paper tries to recreate this hypothetical experiment without the medium of laboratory testing and procedures and only by means of design and publicly available genomic data.

5. Conclusion

Every cell in every organism is intimately connected to the environment surrounding it. Radiation occurs all around us and, depending on the amounts of energy associated with it, can cause significant biological effects observable down to a cellular scale and possibly manifested up to the population level. Over the course of Earth's history, changes to the atmosphere must have played a key role in the conditions under which life formed and evolved (Cnossen et al., 2007). Events in the history of life do very unlikely represent a pure coincidence, but the extent to which they can be linked to the establishment of a modern atmosphere and the fluctuating levels of radiation that came thereafter is not entirely settled (Hessen, 2008).

While it is difficult to quantify the role of naturally-occurring radiation in shaping the outcome of living systems over the course of extended evolutionary time, the results do show a distinct correlation between higher and lower degrees of genetic variance among two different groups of organisms that reside on opposite ends of the radioresistance spectrum; where the order, group or population that has shown increased hypersensitivity to radiation also has an increased rate of molecular evolution due to a higher mutation rate. With that said, the reader should be reminded that correlation does not necessarily indicate a causation. Whether or not naturally-occurring radiation is directly or indirectly implicated in creating genetic variance within populations of living organisms is subject to a follow up systematic investigation.

Conflict of interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

Notes

Note 1. Annotations: AB109397.1, KX790791.1, AF159396.1, FR751074.1, AB819920.1, AF520637.1, AY057808.1, AF159397.1, AB033700.1, AJ698872.1, AB125779.1, AB125777.1, KY587424.1, AJ233955.1, AJ233957.1, Z96053.1, EF187792.1, EU349734.1, FR775869.1, JQ823422.1, DQ191487.1, JQ793903.1, KJ592784.1, U87525.1, KY753976.1, KJ592782.1, KY587421.1, KC735127.1, Z96068.1, JN247708.1, AY156582.1, AY156575.1, AY156572.1, AY156574.1, AY156573.1, AY156578.1, AY156577.1, AY156576.1, AY156579.1, KT188295.1, KT188319.1, KT188221.1, KT188328.1, JQ514257.1, JQ514256.1

Note 2. UGENE was used in comparative sequence analysis. The DNA sequences noted above are in FASTA format. They were obtained from the NCBI database archives.

Note 3. Statistical data and dot distributions were calculated using Excel Formulas. The bell graphs were generated on Geo Gebra.

Supplementary material was provided in the form of .fasta, .aln, .nwk, and .xlsx files.

References

Adam-Guillermin, C., Hertal-Aas, T., Oughton, D., Blanchard, L., …, & Horemans, N. (2018). Radiosensitivity and transgenerational effects in non-human species. *Annals of the ICRP*.

Adewoye, A. B., Lindsay, S. J., Dubrova, Y. E., & Hurles, M. E. (2015). The genome-wide effects of ionizing radiation on mutation induction in the mammalian germline. *Nature communications, 6*, 6684.

Allio, R., Donega, S., Galtier, N., & Nabholz, B. (2017). Large variation in the ratio of mitochondrial to nuclear mutation rate across animals: Implications for genetic diversity and the use of mitochondrial DNA as a molecular marker. *Molecular Biology and Evolution, 34*(11), 2762-2772.

Armas, L. F. (2001). Scorpions of the Greater Antilles, with the description of a new troglobitic species (Scorpiones: Diplocentridae), in: Scorpions 2001. In M. G. A. Polis, V. Fet, & P. A. Selden (Eds.), *British Arachnological Society* (pp. 245-253). Burnham Beeches, Bucks, UK.

Barber, R., Plumb, M. A., Boulton, E., Roux, I., & Dubrova, Y. E. (2002). Elevated mutation rates in the germ line of first-and second-generation offspring of irradiated male mice. *Proceedings of the National Academy of Sciences, 99*(10), 6877-6882.

Blanga-Kanfi, S., Miranda, H., Penn, O., Pupko, T., …, & Huchon, D. (2009). Rodent phylogeny revised: Analysis of six nuclear genes from all major rodent clades. *BMC Evolutionary Biology, 9*(1), 71.

Cai, L., & Wang, P. (1995). Induction of a cytogenetic adaptive response in germ cells of irradiated mice with very low-dose rate of chronic γ-irradiation and its biological influence on radiation-induced DNA or chromosomal damage and cell killing in their male offspring. *Mutagenesis, 10*(2), 95-100.

Carlin, J. L. (2011) Mutations Are the Raw Materials of Evolution. *Nature Education Knowledge, 3*(10), 10.

Churakov, G., Sadasivuni, M. K., Rosenbloom, K. R., Huchon, D., …, & Schmitz, J. (2010). Rodent evolution: Back to the root. *Molecular Biology and Evolution, 27*(6), 1315-1326.

Cnossen, I., Sanz-Forcada, J., Favata, F., Witasse, O., …, & Arnold, N. F. (2007). Habitat of early life: Solar X-ray and UV radiation at Earth's surface 4-3.5 billion years ago. *Journal of Geophysical Research: Planets, 112*(E2).

Cordeiro, A. R., Marques, E. K., & Veiga-Neto, A. J. (1973). Radioresistance of a natural population of Drosophila willistoni living in a radioactive environment. *Mutation Research/Fundamental and Molecular Mechanisms of Mutagenesis, 19*(3), 325-329.

Crow, J. F. (2000). The origins, patterns and implications of human spontaneous mutation. *Nature Reviews Genetics, 1*(1), 40.

D'Suze, G. (1990). Escorpiones, características, distri-bución geográfica y comentarios generales. *Emergencias por animales ponzoñosos en las Américas, 65*.

Deryabina, T. G., Kuchmel, S. V., Nagorskaya, L. L., Hinton, T. G., …, & Smith, J. T. (2015). Long-term census data reveal abundant wildlife populations at Chernobyl. *Current Biology, 25*(19), R824-R826.

Douglas, R. H., & Jeffery, G. (2014). The spectral transmission of ocular media suggests ultraviolet sensitivity is widespread among mammals. *Proceedings of the Royal Society of London B: Biological Sciences, 281*(1780), 20132995.

Drake, J. W., Charlesworth, B., Charlesworth, D., & Crow, J. F. (1998). Rates of spontaneous mutation. *Genetics, 148*(4), 1667-1686.

Dunn, C. W., Zapata, F., Munro, C., Siebert, S., & Hejnol, A. (2018). Pairwise comparisons across species are problematic when analyzing functional genomic data. *Proceedings of the National Academy of Sciences,* 201707515.

Ellegren, H., Smith, N. G., & Webster, M. T. (2003). Mutation rate variation in the mammalian genome. *Current opinion in genetics & development, 13*(6), 562-568.

Fabre, P. H., Hautier, L., Dimitrov, D., & Douzery, E. J. (2012). A glimpse on the pattern of rodent diversification: A phylogenetic approach. *BMC evolutionary biology, 12*(1), 88.

Felsenstein, J. (1981). Evolutionary trees from DNA sequences: A maximum likelihood approach. *Journal of molecular evolution, 17*(6), 368-376.

Fuller, N., Lerebours, A., Smith, J. T., & Ford, A. T. (2015). The biological effects of ionising radiation on Crustaceans: A review. *Aquatic Toxicology, 167*, 55-67.

Gaffin, D. D., & Barker, T. N. (2014). Comparison of scorpion behavioral responses to UV under sunset and nighttime irradiances. *Journal of ARACHNOLOGY,* 111-118.

Gantenbein, B., & Eightley, P. D. (2004). Rates of molecular evolution in nuclear genes of east Mediterranean scorpions. *Evolution, 58*(11), 2486-2497.

Hessen, D. O. (2008). Solar radiation and the evolution of life. *Solar Radiation and Human Health,* 123-136.

Hillis, D. M., & Bull, J. J. (1993). An empirical test of bootstrapping as a method for assessing confidence in phylogenetic analysis. *Systematic Biology, 42*(2), 182-192.

Hodgkinson, A., & Eyre-Walker, A. (2011). Variation in the mutation rate across mammalian genomes. *Nature Reviews Genetics, 12*(11), 756.

Hori, M., Shibuya, K., Sato, M., & Saito, Y. (2014). Lethal effects of short-wavelength visible light on insects. *Scientific Reports, 4*, 7383.

Hsia, C. C., Schmitz, A., Lambertz, M., Perry, S. F., & Maina, J. N. (2013). Evolution of air breathing: Oxygen homeostasis and the transitions from water to land and sky. *Comprehensive Physiology, 3*(2), 849-915.

Husemann, M., Schmitt, T., Stathi, I., & Habel, J. C. (2012). Evolution and radiation in the scorpion Buthus elmoutaouakili Lourenco and Qi 2006 (Scorpiones: Buthidae) at the foothills of the Atlas Mountains (North Africa). *Journal of Heredity, 103*(2), 221-229.

Jenkins, D. G., & Ricklefs, R. E. (2011). *Biogeography and ecology: Two views of one world.*

Joiner, M. C. (1994). Induced radioresistance: An overview and historical perspective. *International Journal of Radiation Biology, 65*(1), 79-84.

Kloock, C. T., Kubli, A., & Reynolds, R. (2010). Ultraviolet light detection: A function of scorpion fluorescence. *Journal of Arachnology, 38*(3), 441-445.

Kumar, S., & Subramanian, S. (2002). Mutation rates in mammalian genomes. *Proceedings of the National Academy of Sciences, 99*(2), 803-808.

Lanfear, R., Kokko, H., & Eyre-Walker, A. (2014). Population size and the rate of evolution. *Trends in ecology & evolution, 29*(1), 33-41.

Lartillot, N., Phillips, M. J., & Ronquist, F. (2016). A mixed relaxed clock model. *Phil. Trans. R. Soc. B, 371*(1699), 20150132.

Lassmann, T., & Sonnhammer, E. L. (2005). Kalign-an accurate and fast multiple sequence alignment algorithm. *BMC bioinformatics, 6*(1), 298.

Levin-Zaidman, S., Englander, J., Shimoni, E., Sharma, A. K., …, & Minsky, A. (2003). Ringlike structure of the Deinococcus radiodurans genome: A key to radioresistance? *Science, 299*(5604), 254-256.

Lindsay, S. J., Rahbari, R., Kaplanis, J., Keane, T., & Hurles, M. (2016). *Striking differences in patterns of germline mutation between mice and humans.* bioRxiv, 082297.

Ling, C. C., & Endlich, B. (1989). Radioresistance induced by oncogenic transformation. *Radiation research, 120*(2), 267-279.

Linschooten, J. O., Verhofstad, N., Gutzkow, K., Olsen, A. K., ... & Godschalk, R. W. (2013). Paternal lifestyle as a potential source of germline mutations transmitted to offspring. *The FASEB Journal, 27*(7), 2873-2879.

Lourenco, W. R. (2001). The scorpion families and their geographical distribution. *Journal of Venomous Animals and Toxins, 7*(1), 03-23.

Madronich, S., McKenzie, R. L., Björn, L. O., & Caldwell, M. M. (1998). Changes in biologically active ultraviolet radiation reaching the Earth's surface. *Journal of Photochemistry and Photobiology B: Biology, 46*(1-3), 5-19.

Monadjem, A., Taylor, P. J., Denys, C., & Cotterill, F. P. (2015). *Rodents of sub-Saharan Africa: A biogeographic and taxonomic synthesis.* Walter de Gruyter GmbH & Co KG.

National Research Council. (2006). Committee to Assess Health Risks from Exposure to Low Levels of Ionizing Radiation, Health Risks from Exposure to Low Levels of Ionizing Radiation: BEIR VII Phase 2. Chapter: 4 *Heritable Genetic Effects of Radiation in Human Populations*.

Ninio, M., Privman, E., Pupko, T., & Friedman, N. (2007). Phylogeny reconstruction: Increasing the accuracy of pairwise distance estimation using Bayesian inference of evolutionary rates. *Bioinformatics, 23*(2), e136-e141.

Nöthel, H. (1987). Adaptation of Drosophila melanogaster populations to high mutation pressure: Evolutionary adjustment of mutation rates. *Proceedings of the National Academy of Sciences, 84*(4), 1045-1049.

Pearson, W. R. (2013). Selecting the right similarity-scoring matrix. *Current Protocols in Bioinformatics, 43*(1), 3-5.

Pray, L. (2008). DNA replication and causes of mutation. *Nature Education, 1*(1), 214.

Prendini, L., & Wheeler, W. C. (2005). Scorpion higher phylogeny and classification, taxonomic anarchy, and standards for peer review in online publishing. *Cladistics, 21*(5), 446-494.

Pryke, L. M. (2016). *Scorpion*. Reaktion Books.

Purvis, A. (1995). A modification to Baum and Ragan's method for combining phylogenetic trees. *Systematic Biology, 44*(2), 251-255.

Rahbari, R., Wuster, A., Lindsay, S. J., Hardwick, R. J., ..., & Stratton, M. R. (2016). Timing, rates and spectra of human germline mutation. *Nature genetics, 48*(2), 126.

Ray, C. (2017). *The Mystery of a Scorpion's Glow*. The New York Times Book of Science Questions & Answers. Penguin Random House.

Samson, P. J., & Ragland, K. W. (1977). Ozone and visibility reduction in the Midwest: Evidence for large-scale transport. *Journal of Applied Meteorology, 16*(10), 1101-1106.

Schlager, G., & Dickie, M. M. (1971). Natural mutation rates in the house mouse estimates for five specific loci and dominant mutations. *Mutation Research/Fundamental and Molecular Mechanisms of Mutagenesis, 11*(1), 89-96.

Sharma, P. P., Baker, C. M., Cosgrove, J. G., Johnson, J. E., ..., & Giribet, G. (2018). A revised dated phylogeny of scorpions: Phylogenomic support for ancient divergence of the temperate Gondwanan family Bothriuridae. *Molecular phylogenetics and evolution, 122*, 37-45.

Smithsonian Institution. (2018). *Fluorescence in Scorpions*. The Walter Reed Biosystematics Unit.

Srinivas, C., Kumar, A., Rai, R., Kini, J., & Kumarchandra, R. (2015). Standardization of mean lethal dose (LD50/30) of X-rays using linear accelerator (LINIAC) in albino wistar rat model based on survival analysis studies and hematological parameters. *Research Journal of Pharmaceutical, Biological and Chemical Sciences, 6*(5), 1215-1219.

Takezaki, N., Rzhetsky, A., & Nei, M. (1995). Phylogenetic test of the molecular clock and linearized trees. *Molecular biology and evolution, 12*(5), 823-833.

Tatebe, K., Chmura, S. J., & Connell, P. P. (2017). Severe Radiation Toxicity Associated with a Germline PTEN Mutation. *International Journal of Radiation Oncology Biology Physics, 99*(2), E620.

Tobe, S. S., Kitchener, A. C., & Linacre, A. M. (2010). Reconstructing mammalian phylogenies: A detailed comparison of the cytochrome b and cytochrome oxidase subunit I mitochondrial genes. *PloS one, 5*(11), e14156.

Uchimura, A., Higuchi, M., Minakuchi, Y., Ohno, M., ..., & Yagi, T. (2015). *Germline mutation rates and the long-term phenotypic effects of mutation accumulation in wild-type laboratory mice and mutator mice*. Genome research.

Upton, A. C., Shore, R. E., & Harley, N. H. (1992). The health effects of low-level ionizing radiation. *Annual Review of Public Health, 13*(1), 127-150.

Verhofstad, N., Linschooten, J. O., van Benthem, J., Dubrova, Y. E., …, & Godschalk, R. W. (2008). New methods for assessing male germ line mutations in humans and genetic risks in their offspring. *Mutagenesis, 23*(4), 241-247.

Weinreich, D. M. (2001). The rates of molecular evolution in rodent and primate mitochondrial DNA. *Journal of Molecular Evolution, 52*(1), 40-50.

Yang, A., Dick, C. W., Yao, X., & Huang, H. (2016). Impacts of biogeographic history and marginal population genetics on species range limits: A case study of Liriodendron chinense. *Scientific reports, 6*, 25632.

Yang, Z. (2006). *Computational Molecular Evolution*. Oxford University Press.

Zepp, R. G., Erickson Iii, D. J., Paul, N. D., & Sulzberger, B. (2007). Interactive effects of solar UV radiation and climate change on biogeochemical cycling. *Photochemical & Photobiological Sciences, 6*(3), 286-300.