Current Debates in Biology:

Classical vs. Molecular Phylogenies

With Regards to Phylogenies of Reptiles.

Students:

Nguyen, Hoang Anh (1700362)

Dumont, Louis (1764955)

November 6, 2018 at 2:30PM

General Biology I (101-NYA-05) sec. 00007

Fall 2018 semester

Date of Submission:

November 20, 2018

Abstract

In this lab, two phylogenetic approaches were observed, the classical and molecular, these distinct methods created a heated debate among the phylogenic field. This debate endued a separation of opinions to question the efficiency of each approach. Our experiment aimed to aid in deciding which would be the most beneficial for a specific group (reptiles). Upon obtaining the results, we concluded that the molecular phylogenetic tree is inferior due to its exorbitant placement of grouping an outgroup species with species that are inherently different and vice versa. To help us decide, we specifically applied both methods to 10 Reptiles species that were universally observed with characteristics ranging from barely different to vastly different, e.g. turtle vs. turtle and turtle vs. snakes, and one outgroup species of Mammals.

Introduction

To begin, phylogenetic research is the area where the fundamental goal is to classify species using the genetic connection and/or relations between them to discover the history of organisms (Hillis, 2003)[1]. The classical phylogeny is an "old" technique that uses physical traits between species to classify them, such as color, number of legs, etc. (Hillis 2003) Whereas molecular phylogeny uses genetic information by looking at short DNA sequences to form a correlation to classify species (P.D.N. Herbert et al., 2005)[2]. Both techniques contribute to the understanding and classification of species. However, which one could lead to a greater understanding of the evolution of species? On one end, DNA barcoding allows an easier, faster and more "modern" way to classify species and gives access to knowledge otherwise inaccessible by the classical method. However, molecular phylogeny offers information that would not be applicable without the long enduring labor of taxonomists[3] (M.C Ebach et al., 2005). Moreover, one could argue barcoding gives little to no information about extinct species, at-risk species or fossils (Hillis, 2003). For extinct species, the classical approach is beneficial due to the lack of access to the DNA sequences and extracting the molecular information demands preserved material. DNA barcoding may be performed well on fossils, but it could potentially destroy the integrity of the samples. Furthermore, extracting the material necessary to classify at-risk species could also endanger them (Hillis, 2003).The goal of this lab was to determine whether the classical or molecular approach would portray better results for the history and representation of the reptiles. This was accomplished by building phylogenetic trees from 10 Reptile species and 1 Mammal outgroup, using both classical and molecular approach.It was determined that the classical approach would be the most beneficial for this experiment due to the fact that molecular approach disregards adaptations of species to their environment and the possibility of climate change affecting this adaptation, therefore the morphological approach would generate the optimal information.

---

[1] Hillis, David. (2003). Molecular Versus Morphological Approaches to Systematics. Annual Review of Ecology and Systematics. 18. 23-42. 10.1146/annurev.es.18.110187.000323.
[2] Hebert, P.D.N., and Gregory, T.R. (2005). The promise of DNA barcoding for taxonomy. Systematic Biology 54:852-859.
[3] Ebach, M.C. and Holdrege, C. (2005). More taxonomy, not DNA barcoding. Bioscience 55: 822-823

Materials and Methods

For our classical phylogenetic tree, we worked with the Reptiles class, specifically with the following species: *Common Snapping Turtle, Western Pond Turtle, Spiny Softshell Turtle, Caiman Crocodile, Nile Crocodile, Galápagos Land Iguana, Tokay Gecko, King cobra, Western Diamondback rattlesnake, Black Neck Garter Snake, and Bighorn Sheep (outgroup).* Amongst these species, we selected the following characters to build the tree: venomous, lives in aquatic environment, cold-blooded, carnivorous, four-chambered heart, have hemipenes, is diurnal, shed scales in smaller pieces, has a carapace, has ears. As for our molecular phylogenetic tree, we utilized Nucleotide BLAST app from the National Center for Biotechnology Information[4]. We used the outgroup species Bighorn sheep DNA sequence as a query and the sequences of the listed Reptile species above as subjects[5]. For BLAST algorithm, we chose 'somewhat similar' (blastn) because the species were not closely similar enough ($> 85\%$).

---

[4] Nucleotide BLAST Web Application. https://blast.ncbi.nlm.nih.gov/Blast.cgi
[5] DNA sequences courtesy of BoldSystems. http://www.boldsystems.org/index.php

Results

Table 1 – Morphological Differences

	A	B	C	D	E	F	G	H	I	J	X
A		1	1	3	3	4	5	8	7	7	6
B			0	4	4	3	6	7	7	6	6
C				4	4	3	6	7	7	6	6
D					0	5	4	7	7	6	5
E						5	4	7	7	6	5
F							3	4	4	3	4
G								3	1	2	6
H									0	1	6
I										1	6
J											5
K											

Legend:

Common Snapping Turtle	A	**Nile Crocodile**	E	**Western Diamondback Rattlesnake**	I
Western Pond Turtle	B	**Galapagos Land Iguana**	F	**Black Neck Garter Snake**	J
Spiny Softshell Turtle	C	**Tokay Gecko**	G	**Bighorn Sheep**	X
Caiman Crocodile	D	**King Cobra**	H		

From Table 1, we observe that the Turtle species group (A, B, C) has at least 5 to 8 differences from the Snake and Cobra species group (H, I, J). On the other hand, the Turtle species group has, on average, fewer differences than the Crocodile (D, E) and Iguana (F) species group. A noticeable intermediary species can be seen as the Tokay Gecko (G) as it has 0 to 3 differences to Snake and Cobra group whilst having no lower than 3 differences to Turtle, Crocodile and Iguana group.
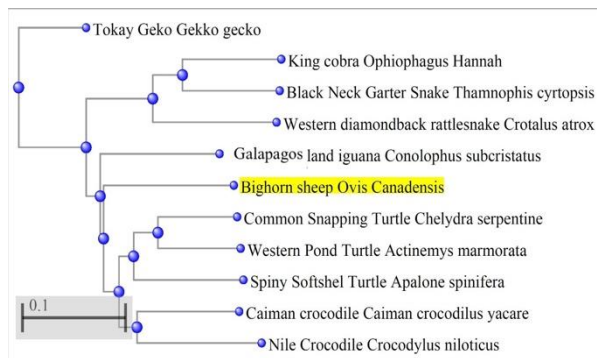
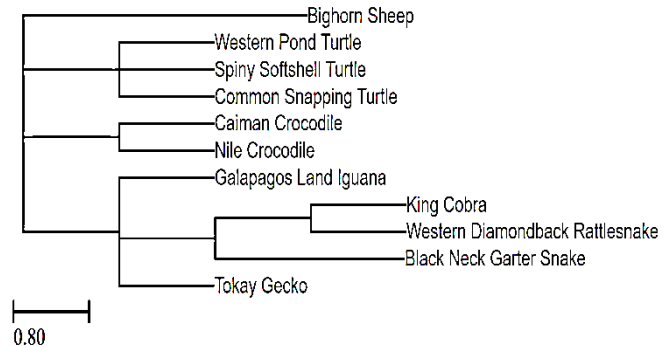*Fig. 1 – Molecular Phylogenetic Tree based on Nucleotides BLAST by NCBI.*

Fig. 2 – Morphological Phylogenetic Tree based on Data Matrix and Table of Differences

From Figure 1, the molecular phylogenetic tree, we can observe that the outgroup (Bighorn sheep Ovis Canadensis) is located near the middle of the tree, effectively placing a mammal among many other reptiles. Also from Figure 1, the Tokay Gecko (Gekko Gecko) is placed outermost from the Reptiles. Figure 2 (morphological phylogenetic tree) tells a different story where the Tokay Gecko is placed in the same groupings of Reptiles and the Bighorn Sheep outgroup is placed expectedly outermost of the phylogenetic tree.

Discussion and Conclusion

Our results show that there is one significant difference between the morphological and molecular phylogenetic tree which is the placement of our outgroup, i.e. *Bighorn Sheep.* The molecular phylogeny seems rather interesting, placing a sheep amongst turtles, iguanas and crocodiles (see fig. 1) whilst, objectively speaking, a sheep is visually different from the others. Upon that initial observation of the results, we are inclined to support the method of classic phylogeny.

Our expectation for the results was the molecular phylogenetic tree would only differ from the classic phylogenetic tree by a small margin, a margin that we could understand, interpret and justify. While the molecular placement of groups of species is relatively logical, e.g. snakes with cobras or different species of turtles together, but completely misses the mark when it comes to placing the Bighorn Sheep – a mammal, a species outside of the Reptiles class – against the realistic perception. The ridiculousness of the results obtained from the molecular phylogenetic tree is complacent with the audacious claims from Hebert that DNA barcoding could be the primary factor driving today's taxonomical researches (Hebert,

2005:853). How can one look at an iguana and confidently say that it reminds him/her of a sheep? For sure, with the molecular approach of DNA barcoding, we can quickly and accurately obtain a plethora of "exciting" new information, but that does not necessarily mean deceiving our perception of the natural world where biodiversity is certainly visible and differentiable.

Pivoting our observation onto another species and again we see the same illogical placement of a Reptile, the Tokay Gecko, furthermost away from the species within its class. So, we raise an important question: *did we classify species wrongly from the birth of modern taxonomy or DNA barcoding is just another application of technology propelled by Web 3.0-engrossed taxonomists?* For us, it is the latter. Because when we look at a species and try to make sense of it, we need to holistically view how the species looks visually, the habitats in which it grows and reproduces comparing to the neighboring species. We cannot systematically look at computer-interpreted DNA barcodes and being unable to relate such information to the existing scientific body of knowledge (Will et al., 2005:846)

References

Ebach, M.C. and Holdrege, C. (2005). More taxonomy, not DNA barcoding.

    Bioscience 55: 822-823

Hillis, David. (2003). Molecular Versus Morphological Approaches to Systematics.

    Annual Review of Ecology and Systematics. 18. 23-42.

    10.1146/annurev.es.18.110187.000323.

Hebert, P.D.N., and Gregory, T.R. (2005). The promise of DNA barcoding for taxonomy.

    Systematic Biology 54:852-859.

Will, Kipling & D Mishler, Brent & Wheeler, Quentin. (2005). The Perils of DNA Barcoding

    and the Need for Integrative Taxonomy. Systematic biology. 54. 844-51.
    10.1080/10635150500354878.