

A possible case of adaptive radiation in cycads. A commentary on ‘Transcriptome sequencing data provide a solid base to understand the phylogenetic relationships, biogeography and reticulated evolution of the genus *Zamia* L. (Cycadales: Zamiaceae)’

José Said Gutiérrez-Ortega^{1,*}
and Juan Carlos Villarreal A.^{2,*}

¹RIKEN Interdisciplinary Theoretical and Mathematical Sciences Program, Wako, Saitama 351-0198, Japan and

²Department of Biology, Université Laval, Québec, G1V 0A6, Canada

*For correspondence. E-mail jose.go@riken.jp or jcvil9@ulaval.ca

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Charismatic and endangered, with beautiful yet primitive aspects appreciated by scientists and enthusiasts alike, cycads encompass 375 extant species that place their most common ancestor as far back as the Carboniferous (~330 million years ago; Coiro *et al.*, 2023). However new evidence of evolutionary diversification among cycads has challenged the well-worn tag of ‘living fossil’ (Coiro and Seyfullah, 2024). Indeed 375 species is not a small number for a gymnosperm order, suggesting that cycads met multiple evolutionary opportunities that promoted diversification and persistence. It also suggests resilience during climate change events and adaptability to new environmental conditions, as exemplified by the genus *Dioon* (Gutiérrez-Ortega *et al.*, 2024). Of the ten extant cycad genera, *Cycas* and *Zamia* comprise more than half of the total cycad species in the world and the best examples of the cycads’ success. Both genera have been the focus of intensive studies to try to understand the geological and evolutionary factors that have produced such spectacular diversification in the Cenozoic (e.g. Liu *et al.*, 2018; Calonje *et al.*, 2019).

The landmark study by Nagalingum *et al.* (2011) presented evidence that the extant cycad genera experienced a global synchronous radiation, as estimated

through phylogenetic tree reconstructions based on one nuclear and two organellar markers. The study prompted a plethora of papers dealing with cycad diversity (e.g. Condamine *et al.*, 2015; Liu *et al.*, 2018). However nearly all of them faced a similar problem: that the few loci used lack enough variation to tease apart the infra-generic relationships in the most species-rich genera, especially in *Cycas* and *Zamia*. Despite the shortcomings, Calonje *et al.* (2019) provided a complete account of the genus *Zamia* using a wide array of dating, biogeographical and diversification approaches from one plastid and nine nuclear markers.

The study by Lindstrom *et al.* (2024) in this issue of *Annals of Botany* takes the study by Calonje *et al.* (2019) to a completely different level in terms of the amount of analysed data. Lindstrom *et al.* (2024) used more than 2900 loci recovered from the transcriptomes of 77 species (~90 % of the extant *Zamia* species). They used coalescent and network approaches to unravel the evolutionary history of the genus, including species relationships and diversification analyses.

Lindstrom *et al.* (2024) not only confirm the finding of Calonje *et al.* (2019), but also extend our understanding of the biogeography of the Neotropics by demonstrating a clear correspondence between the clade delimitations and the geographical region to which they belong. For example, Lindstrom *et al.* (2024) found that a clade delimited to the Caribbean region (clade I) is sister to a clade that includes three species from eastern Mexico (clade II). The major clade I + II is in turn sister to all remaining clades: clade III includes most species from Mesoamerica, clade IV is represented by a single species (*Z. soconuscensis*), clade V by several species from the Isthmus of Panama region, and clades VI and VII comprise all species from South America. All these clades (crown ages) diversified between 8 and 20 million years ago, suggesting a highly dynamic speciation process in the Late Miocene. By using phylogenetic network analyses, the authors uncovered three instances of ancient reticulation events in the evolutionary history of *Zamia*, in the clade of *Z. fisheri* and the ancestor of *Z. soconuscensis*. Because evidence of hybridization in wild cycads is almost in-existent (but see Gutiérrez-Ortega *et al.*

et al., 2023), this unexpected finding sets the ground to explore the consequences of hybridization in cycads while posing challenges in the phylogenetic reconstruction.

The findings of Lindstrom *et al.* (2024) represent the most comprehensive phylogenetic reconstruction of the genus *Zamia* to date. But does this research represent the definitive phylogenetic reconstruction of the genus? Not necessarily. Lindstrom *et al.* (2024) still falls over with respect to common problems inherent within methodological choices taken in phylogenomic research, such as phylogenetic uncertainty, the impact of incomplete lineage sorting, the anomaly zone and ancient reticulation in phylogenies. However, the authors thoroughly explored the problem of how different assumptions about the dataset will inevitably provide discordant trees. For example, they played with two variables: (1) the exclusion/inclusion of the third codon position in their dataset, and (2) consideration of the whole dataset as a single partition (all loci concatenated) or as multiple partitions (each locus being independent). By combining the two variants of the two variables, we obtained a total of four trees. The authors showed that none of the four trees are completely identical, as there are discordances either in the positions of clade II or in the compositions of clades VI and VII. At the end, for the sake of coherency, and under the assumption that all gene trees might have the same weight in evolution, the authors chose the tree obtained with the combination that is ‘excluding the third codon and considering multiple partitions’, on which they based all subsequent analyses. We understand that it is virtually impossible to decide which of the four methodological choices is the most accurate due to the fact that phylogenetic reconstructions at the time scale in the order of millions of years are not testable or falsifiable (Fitzhugh, 2016). The study is a reminder that the search for a bifurcating species tree may be illusory and network approaches may represent a more accurate depiction of diverse genealogies in plants.

In addition, one aspect that took our attention is the possibility that the diversification of the genus *Zamia* can be explained using the concept of adaptive radiation. Adaptive radiation refers to the



FIG. 1. Among all the cycad genera, *Zamia* displays the most spectacular diversity in habitats and morphology. (A) A good example is the only epiphytic gymnosperm, *Z. pseudoparasitica*, as shown climbing up to 20 m in the Panamanian rainforest canopy (photograph by Lilisbeth Rodriguez-Castro and Mia Roy). (B) The natural history of this and other *Zamia* species is being unravelled, as shown by the recently discovered seed disperser of *Z. pseudoparasitica*: the nocturnal northern olingo (*Bassaricyon gabbii*) is seen approaching a seed cone (photograph by Claudio Monteza-Moreno and Lilisbeth Rodriguez-Castro).

disparate evolution of phenotypic and ecological diversity in a fast-evolving lineage (Schluter, 2000). The study showed that *Zamia* diversification might have been in constant decline since its single peak at the origin of the genus (~29 million years ago), indicating its fast initial evolution. Also, while all *Zamia* species are also relatively easy to recognize based on vegetative and reproductive traits (an obvious feature denoting phenotypic diversity), they are also distinct by occupying a widely varied set of habitats throughout the heterogeneous American continent. Some good examples of the disparate morphological and functional evolution of the genus are the only known epiphytic gymnosperm, *Z. pseudoparasitica* from Panama (Bell-Doyon and Villarreal, 2020) (Fig. 1), *Z. roezlii*, being able to live in swamps and mangroves in Colombia, or the smallest cycad in the world, *Z. pygmaea* (up to 25 cm) from Cuba. Their respective unique

characteristics do not seem to respond to a series of adaptive traits gained through synapomorphies, but to phylogenetically independent adaptations gained through their demographic histories under specific environmental pressures. This may also explain the high homoplasy of phenotypic characters in the genus. If *Zamia* represents indeed a case of adaptive radiation, it raises important questions regarding the apparently disparate mechanisms that triggered their speciation, and what key innovations have helped them to persist to date.

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