

Evolutionary Inequality: Comparing Phylogenetic Relationships with Other Biological Properties

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Summary

Molecular methods enable more reliable reconstructions of phylogenetic relationships than a strictly morphological approach, making increasingly clear that the tree of life as a well-balanced structure, is a misleading metaphor. This is illustrated with some case histories of land snails' phylogenies. Prime species and remnant species are defined and discussed. Reproductive isolation cannot always be concluded from only molecular data. Therefore, genetic barcoding is not unreservedly applicable in all cases. Single-gene speciation in snails may illustrate this most convincingly.

Introduction

Inequality is a basic characteristic of the evolutionary process. The common ancestor of the chordates, for instance, has developed into sea squirts (ascidians) in one lineage and hominids in another, showing that differentiation may take place at quite different rates. Comparably unequal may be the amount of speciation and/or extinction in sister-groups, which may additionally show extreme differences in the size of their ranges. All this became increasingly clear with the rise of phylogenetic systematics and even more so by the introduction of molecular methods. The undisputedly great value of DNA sequencing should not be overvalued however. Phylogenetic relationships and taxonomic status may diverge and reproductive isolation, which is decisive in certain species definitions, cannot always be derived from only molecular data.

Among species of gastropod molluscs evolutionary incongruence is illustrated by both 'remnant' species, representing single lineages with speciose sister-groups, and

'prime' species, which largely and sometimes misleadingly determine the image of a higher taxon. The fact that taxa that are most closely related, may be reproductively isolated, whereas more distantly related taxa within the same genus may hybridize freely, is illustrated. Since single-gene speciation is possible in snails, species may occur that are genetically extremely similar but nevertheless separate by pre-mating isolation. Obviously, such species cannot be recognized by modern genetic barcoding.

Materials and Methods

On the basis of existing molecular phylogeny reconstructions and biogeographic patterns acquired by fieldwork, general conclusions on the nature of species are presented.

Results

Prime species and remnant species

In the helicid genus *Arianta* Turton, 1831, *A. arbustorum* is exceptional because of its very large range, which is not restricted to mountainous areas, where the far more locally distributed, congeneric taxa are found. It occurs with several subspecies in the Alps, the Pyrenees and central Europe, including southern Scandinavia and Britain [1]. In *Theba* Risso, 1826, another genus of the Helicidae, 17 species or subspecies are actually known, all of which restricted to Morocco, southernmost Spain

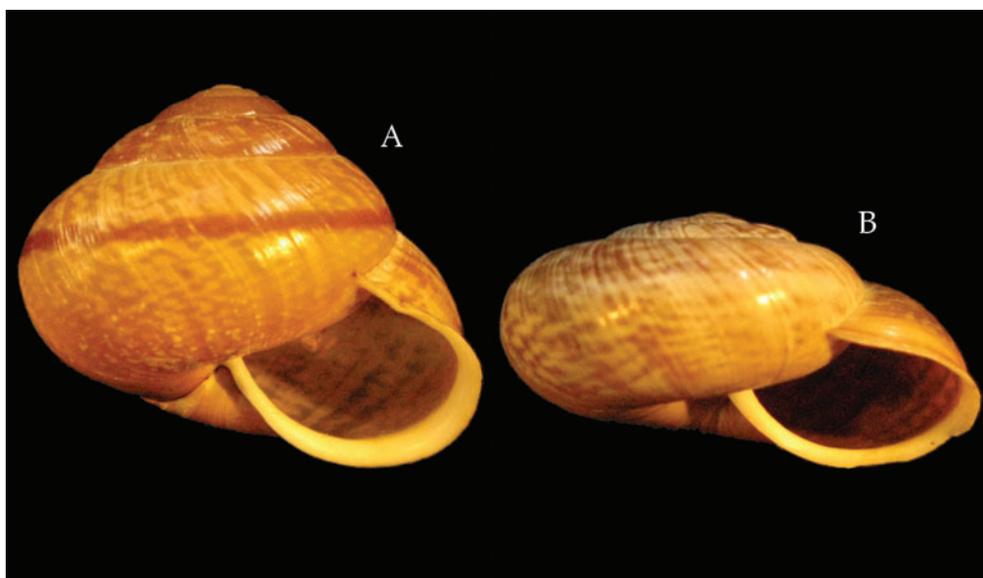


Figure 1. A *Arianta arbustorum arbustorum*, 'prime' species and B *A. schmidtii*, 'remnant' species in *Arianta*.

and the eastern Canary islands [2]. Only a single species, in fact a subspecies, i.e. *Theba pisana pisana* (L., 1758), occurs in the entire Mediterranean region and along the Atlantic coasts from Morocco to Belgium, SW. England and E. Ireland [3]. By their common occurrence and very wide distributional areas, *Arianta arbustorum* (Fig. 1A) and *Theba pisana* exemplify the concept of prime species. Similarly, *Abida secale secale* (Draparnaud, 1801) is the prime taxon in *Abida* Turton, 1831, because of its exceptionally large range, extending from the British isles to Austria, with a single locality in Slovakia [4, 5]. The ten other *Abida* species occupy much smaller ranges in the Pyrenees and the Cantabrian mountains, with only *A. polyodon* (Draparnaud, 1801) extending from the Pyrenees into the Mediterranean zone of southern France and southwards into eastern Spain [6]. In *Orcula* Held, 1838, the prime species is *O. dolium* (Draparnaud, 1801), occurring in the Alps and the Carpathians, in contrast to the c. ten congeneric species, which have much smaller ranges in southeastern Europe [3, 7]. In *Chondrina* Reichenbach, 1828, it is *C. avenacea* (Bruguière, 1792) (Figs 3, 4), occurring in eastern Spain, central Europe, Italy, and the NW. Balkans [4, 8], with *C. arcadica* (Reinhardt, 1881) reaching from eastern Switzerland and (isolated) southern Scandinavia into Asia Minor [8].

There are molecular phylogeny reconstructions for *Arianta*, *Chondrina*, and *Abida*,

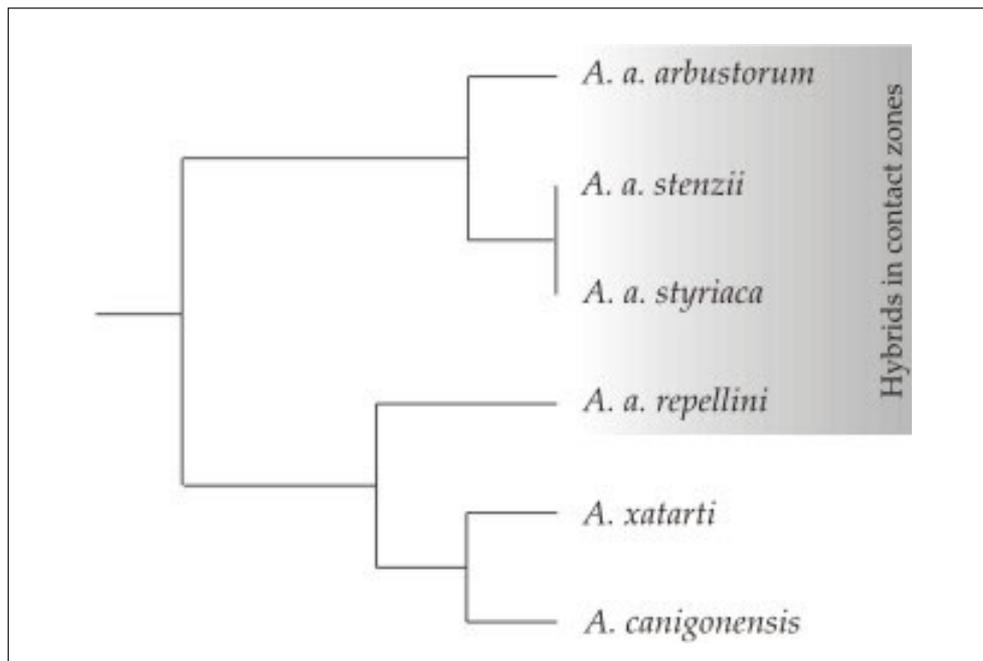


Figure 2. Phylogenetic relationships among some *Arianta* species and subspecies (after Gittenberger et al., 2004). Only *A. xatarti* and *A. canigonensis* are reproductively isolated; the 4 other taxa are interconnected by hybrid zones.

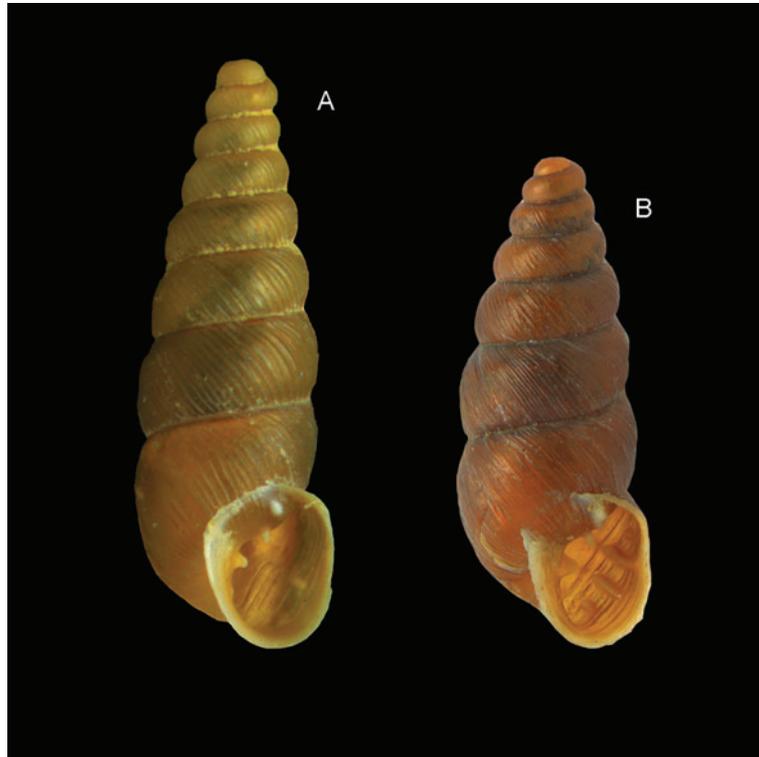


Figure 3. A *Chondrina avenacea*, 'prime' species, and B *C. maginensis*, 'remnant' species in *Chondrina*.

but not for *Theba*, and *Orcula*. In *Arianta*, *A. schmidtii* (Rossmässler, 1836) (Fig. 1B) turned out to be the sister group of the entire *A. arbustorum* complex, which is distributed with several subspecies in the Alps, the Pyrenees and central Europe, including southern Scandinavia and Britain [1]. It is a poorly known, high-alpine, remnant species, restricted to a few mountain peaks in the extreme southeastern part of the Alps in Austria, where it is most probably extinct now [9], and Slovenia [10].

A molecular phylogeny reconstruction for *Chondrina* (Fig. 5) resulted in the discovery of an even more extreme example of the remnant species concept. *Chondrina* contains about 26 species, occurring throughout the mountainous parts of central and southern Europe, Asia Minor and N. Africa [6]. The snails occur nearly exclusively on exposed, vertical limestone cliffs. In that habitat a species is often found in great numbers, whereas no more than two species may coexist. The radiation in *Chondrina* is largely nonadaptive, as defined by Gittenberger [11, 12]. The genus is most speciose in southwestern Europe, the Iberian peninsula, including the French part of the Pyrenees and Morocco. In the Iberian peninsula limestone-islands with *Chondrina* are found in the more or less isolated sierra's, where 14 species are currently recognized.

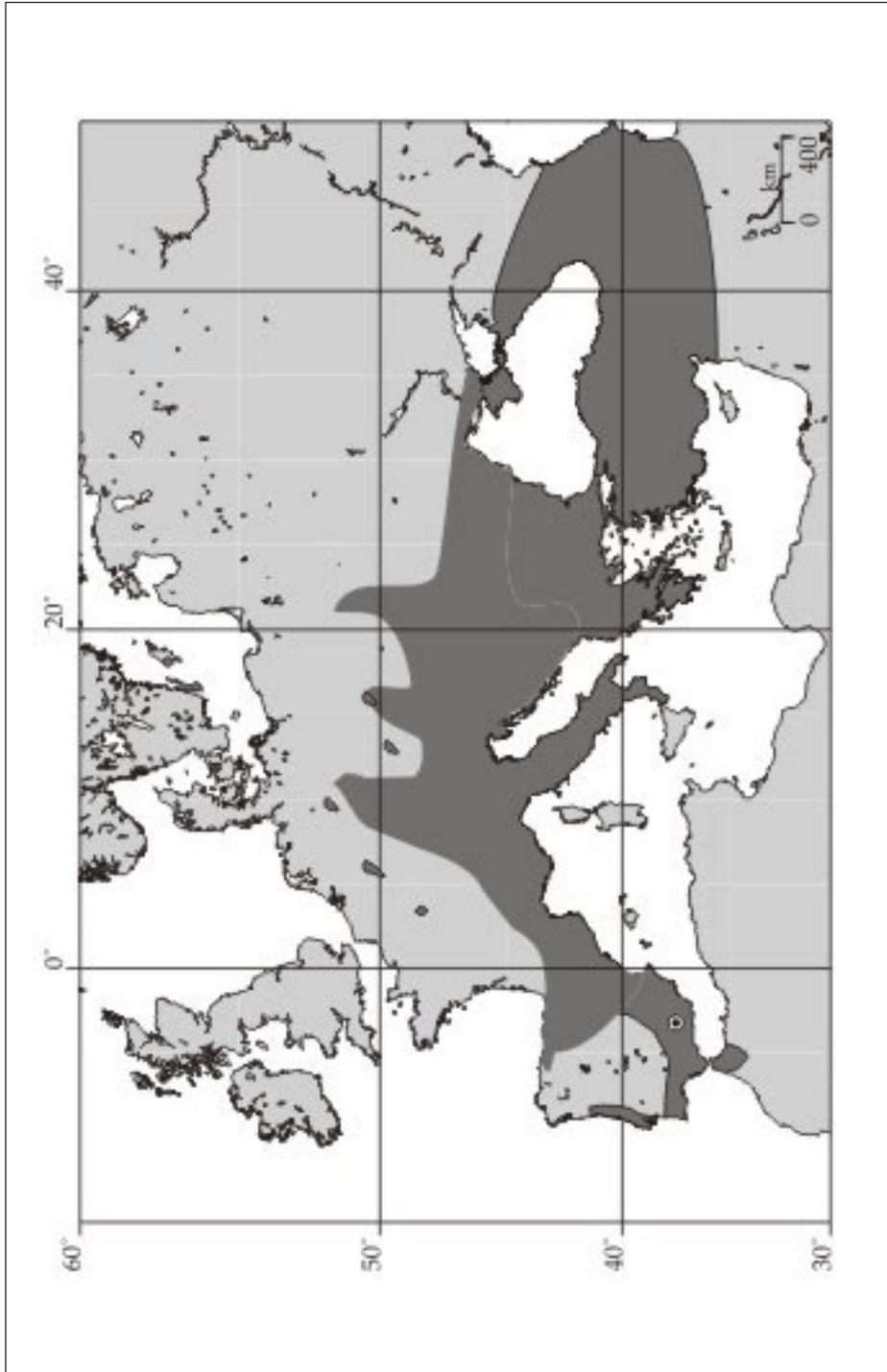


Figure 4. Geographical map with records for *Chondrina maginensis* and the total range of the genus *Chondrina* (after Kokshoorn, 2008 [thesis]).

Three more species are known from Morocco, so that more than half of all known species of *Chondrina* are concentrated in SW. Europe and Morocco.

Surprisingly, the DNA analysis made clear that *Chondrina maginensis* Arrébola & Gómez, 1998, has to be considered the sistergroup of all the other extant *Chondrina* species that could be investigated (Fig. 5). *Chondrina maginensis* is known from the Spanish department of Jaén, where it occurs at only a few localities in the Sierra Mágina (c. 25 km W. of the city of Jaén). The generic classification of this remnant species, which is somewhat aberrant by its slender conical shell shape, could be confirmed by studying the radula.

For the genus *Abida*, a comparable remnant species cannot be indicated, since none of the species has an extremely small range. In both *Theba* and *Orcula*, species with very small ranges are known, but since there is no phylogeny reconstruction available for these genera, we can only speculate that one or more of these narrowly endemic species could be considered a remnant taxon.

The origin of prime and remnant species

To enable an in depth discussion about the evolutionary origin of prime and remnant species, data on a much higher number of genera with strongly contrasting species' ranges, are necessary. Recent research on species' introductions might be relevant here, since world-wide invasive species can be seen as a special category of prime species.

In *Arianta*, *Chondrina* and *Abida*, the prime species evolved relatively recently, whereas the lineages of remnant species are older. *Arianta arbustorum arbustorum* might have reached its actual, large range because of an ecological 'key innovation' which enabled it to occur in the lowlands, in contrast to the congeneric taxa, which remained dependent on an alpine habitat. This might not apply to the *Chondrina* and *Abida* prime species, occurring in habitats similar to those of their congeneric relatives. Maybe, these prime species achieved their status by chance. After an accidental, successful, dispersal event, closely related species might not be able anymore to colonize the same area because of the competitive exclusion principle (Gause's Law). The fact that the prime species originated relatively recently suggests that their status is only temporary. However, if *Solatopupa similis* is considered the prime species in *Solatopupa*, we have to conclude that prime species may be relatively old elements as well [13]. For the moment being, prime species are best defined on the basis of only their evolutionary success, i.e. the relative size of their geographic ranges. To be called a remnant species, an additional criterion is necessary. The phylogenetic relationships should be known to differentiate between a primary and a secondary situation. Only in the latter case the term remnant would be appropriate.

Phylogenetic relationships, reproductive isolation and genetic barcoding

The molecular phylogeny reconstruction of *Arianta* taxa (Fig. 2) made clear that phylogenetic relationships cannot unequivocally be used to conclude about reproductive isolation, i.e. speciation. Obviously, the narrowly endemic taxa *Arianta xatarti* and *A. canigonensis*, which are restricted nowadays to some or even only a single

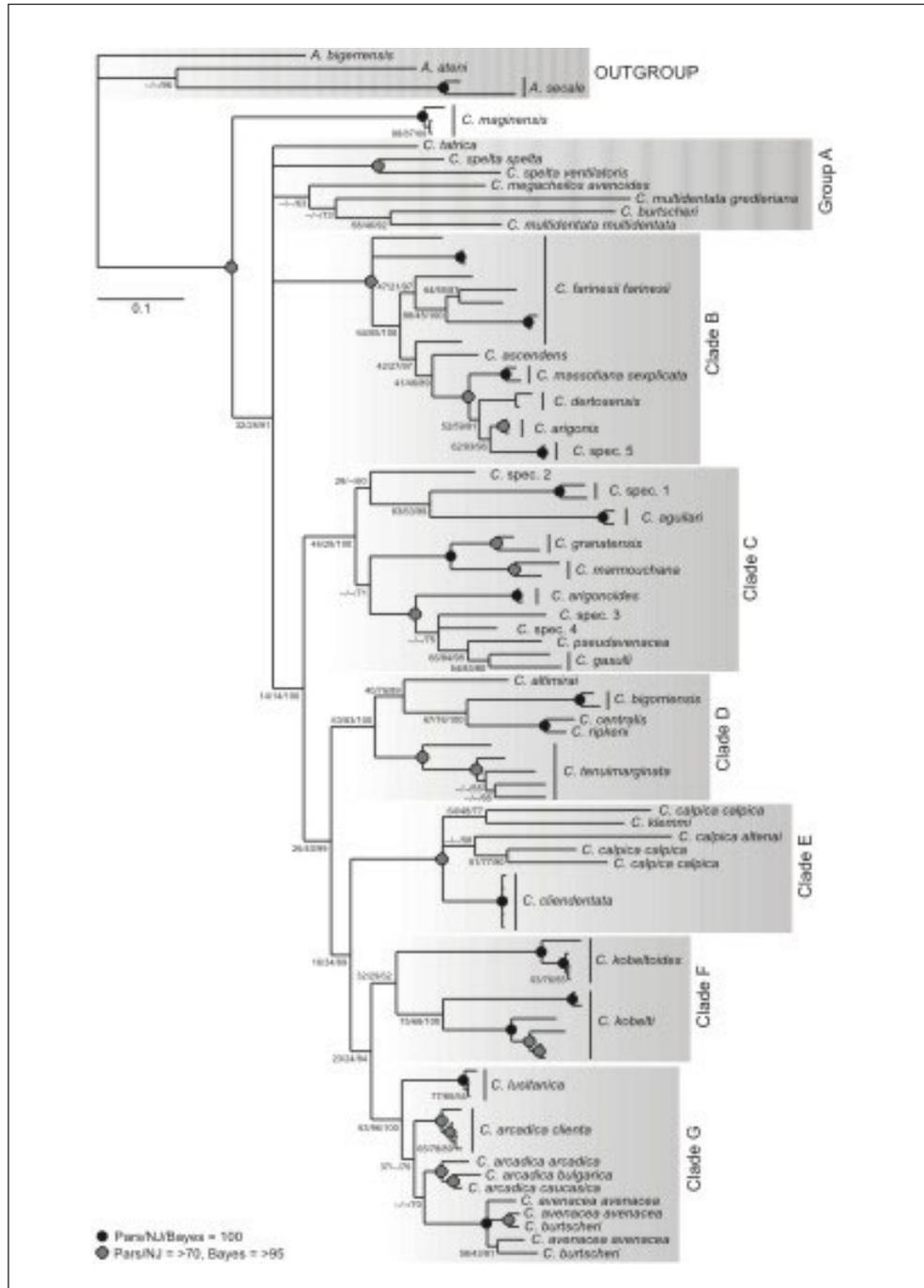


Figure 5. Molecular phylogeny reconstruction for the *Chondrina* species (after Kokshoorn, 2008 [thesis]).

Pyrenean mountain peak, have been under a much more severe selection pressure and bottleneck-effect than the other *Arianta* taxa, among which *A. a. arbustorum* survived the Pleistocene glaciations in large refugia.

Species recognition on the basis of only molecular data, i.e. genetic barcoding, remains problematic since the DNA used for the molecular diagnosis is not related to the gene or genes that are responsible for reproductive isolation. This is most obvious in cases of single-gene speciation in snails [14, 15, 16]. In the early stages of this unique speciation mechanism, there may be complete pre-mating isolation but hardly any genetic differentiation.

Conclusions

Data from DNA sequencing may be indispensable for the most reliable phylogeny reconstruction and historical biogeography. By their application it turned out that widespread, common, 'prime' species may be 'young' species, dating from a relatively recent speciation event. Maybe such species exemplify a temporary phase in evolution. It remains unclear whether a key innovation or coincidence, or both, are basic to their evolutionary success. Narrowly endemic, 'remnant' species may be 'old' species, dating from a relatively early speciation event.

Molecular methods are not always conclusive when the species status is disputed. Sister groups may be reproductively isolated and less closely related forms may hybridize freely. Therefore, reproductive isolation cannot simply be derived from the phylogenetic relationships [1, 17]. Among snails, single-gene speciation may result in total reproductive isolation because of a mutation in the chirality gene. In an early phase of speciation based on this mechanism, the evolving species cannot be diagnosed by genetic barcoding.

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