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Supplementary data

"Data Supplement"

References

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Evolutionary biology

Forest refugia in Western and Central Africa as ‘museums’ of Mesozoic biodiversity

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2CNRS, Université Paul Sabatier, ENFA, UMR 5174 EDB (Laboratoire Évolution et Diversité Biologique), Université de Toulouse, 118 route de Narbonne, 31062 Toulouse, France
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The refugial speciation model, or ‘species pump’, is widely accepted in the context of tropical biogeography and has been advocated as an explanation for present species distributions in tropical Western and Central Africa. In order to test this hypothesis, a phylogeny of the cryptic arachnid order Ricinulei, based on four nuclear and mitochondrial DNA markers, was inferred. This ancient clade of litter-dwelling arthropods, endemic to the primary forests of Western and Central Africa and the Neotropics, might provide insights into the mode and tempo of evolution in Africa. Twenty-six African ricinuleid specimens were sampled from eight countries spanning the distribution of Ricinulei on the continent, and analysed together with Neotropical samples plus other arachnid outgroups. The phylogenetic and molecular dating results suggest that Ricinulei diversified in association with the fragmentation of Gondwana. The early diversification of Ricinoides in Western and Central Africa around 88 (±33) Ma fits old palaeogeographical events better than recent climatic fluctuations. Unlike most recent molecular studies, these results agree with fossil evidence, suggesting that refugia may have acted as ‘museums’ conserving ancient diversity rather than as engines generating diversity during successive episodes of climatic fluctuation in Africa.

1. Introduction

The existence of forest refugia is broadly accepted in tropical biogeography, because there is ample evidence for forest fragmentation linked to past climatic change in temperate and tropical regions [1,2]. Two different hypotheses have been advanced regarding the role of refugia in shaping the current biodiversity of tropical Africa. The refugial speciation model [3], or ‘species pump’, invokes genetic differentiation between allopatric populations, fragmented and trapped in refugia by the expansion of savannah during Quaternary glacial maxima [4]. This hypothesis predicts the origin of most rainforest species to be relatively recent [5]. On the other hand, past climatic changes could have depleted rather than augmented biodiversity, suggesting that refugia might have acted as ‘museums’ conserving ancient lineages, and divergence between sister species might predate climatic fluctuations [6]. Whereas fossil evidence for plants has long suggested an ancient diversity followed by cycles of extinction owing to climatic fluctuation [7], most molecular studies (see the electronic supplementary material,
Table 1. Tissue sample numbers, provenance data, GenBank accession numbers and voucher repositories for Ricinulei and outgroup taxa from which DNA sequence data were generated for the present study. COI, cytochrome c oxidase subunit I.

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Outgroups

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  - Rhipicephalus sanguineus (Latreille, 1806)
    - L76342
  - Carios capensis (Neumann, 1901)
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  - Amblypygi
    - Phrynus sp.
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  - Araneae
    - Calisoga longitarsis (Simon, 1891)
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    - Hahpepelta schmidt von Wirth, 1991
      - AY425722.1
    - Habronattus arogenensis (Peckham & Peckham, 1888)
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    - Neptila davata L. Koch, 1878
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    - Hypochilus theorellii Marx, 1888
      - AF103505
      - AY425719.1
  - Scorpiones
    - Buthus occitanus (Amareux, 1789)
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    - Centruroides limpidus (Karsch, 1879)
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    - Mesobuthus martensii (Karsch, 1879)
      - F394879.1
    - Uroctanus mordan Thorell, 1876
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  - Solifugae
    - Eremobates palpisetulosus (Fichter, 1941)
      - NC_010779
    - Mastigoproctus giganteus (Lucas, 1835)
      - AY859587.1
    - Phalangium opilio Linnaeus, 1758
      - AF124937
    - Limulus polyphemus (Linnaeus, 1758)
      - L81949

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figure S1) portray climatic fluctuation as an agent of speciation and diversification.

Because of their great age [8], extreme endemism [9,10] and low vagility, Ricinulei Thorell, 1876 represent an excellent model for studying the biogeography of Western and Central Africa. Commonly known as ‘hooded tick spiders’ or ‘tick beetles’, these small (less than 11 mm) predatory arthropods are among the most obscure and cryptic of the arachnid orders [11]. A mere 72 extant ricinuleid species are currently described [12], and grouped in three genera: Ricinoides Ewing, 1929 from tropical Western and Central Africa, and the Neotropical Cryptocellus Westwood, 1874 and Pseudocellus Platnick, 1980. African ricinuleids are restricted to the moist soil and litter habitats of rainforests [13], whereas Neotropical ricinuleids have also been collected in caves [12].

The first molecular phylogeny of Ricinulei is presented here, with the aims of providing a temporal framework for the diversification of Ricinoides in Western and Central Africa, and investigating the effects of forest refugia on the generation and maintenance of tropical biodiversity.

2. Material and methods

Eleven Ricinoides species, recorded from 14 countries (including first record of Ricinulei from Senegal, reported here), are currently recognized in Africa [13,14]. Specimens were collected by sifting leaf litter, Winkler extraction and actively searching in appropriate habitats (especially under logs in forested areas) throughout their known distributional range. Voucher specimens used for DNA isolation have been deposited in the following institutions: American Museum of Natural History (AMNH), New York, NY, USA; Musee Royal de l’Afrique Central (MRAC), Tervuren, Belgium; Museum of Comparative Zoology (MCZ), Harvard University, Cambridge, MA, USA (table 1). The ingroup taxon sample comprises 26 specimens, representing at least seven ricinuleid species, from eight African countries (figure 1). Ten Neotropical ricinuleids and 20 representatives of seven other chelicerate orders were included as outgroup taxa (table 1).

DNA extraction and sequencing were conducted using protocols optimized for other arachnids [16,17] for the following genes: 12S rRNA (12S), cytochrome c oxidase subunit I, 18S rRNA (18S) and 28S rRNA (28S). Sequences were aligned using MUSCLE v. 3.7 [18]. Divergence time estimation was performed in a Bayesian framework using BEAST v. 1.5.4 [19], with an uncorrelated lognormal model of rate evolution [20]. This approach integrates the uncertainty of calibration points and topology, considered important because the phylogenetic placement of Ricinulei within Arachnida remains uncertain [21].

BEAST does not use a coupled Markov chain Monte Carlo (MCMC), potentially making it more prone to becoming trapped in local optima. A maximum-likelihood analysis was, therefore, performed using RAxML HPC v. 7.2.7 [22] with a GTR + gamma model applied to each gene. The optimal tree was then used as an initial starting tree for BEAST. Arachnid diversification was constrained based on the oldest known fossils [23], using an exponential distribution prior and setting the standard deviation to obtain a 95 per cent range between 428 (oldest...
Orthodorus mouhata Acari
Carpos capitensis Acari
Ixodes hexagonus Acari
Haemaphysalis fara Acari
Rhipicephalus sanguineus Acari
Uroctomus norda Scorpionidae
Centruroides limpidus Scorpionidae
Buthus occitanus Scorpionidae
Mesobuthus martensii Scorpionidae
Mastigoproctus giganteus Thelyphonida
Hephaethula hangzhouensis Araneae
Hypochilus thorelli Aranae
Calisoga longitarsis Araneae
Ornithoctonus huventa Araneae
Nepilida clavata Araneae
Habronattus oregonensis Araneae
Phadangium opilio Opilionidae
Phrynus sp. Amblypygi
Eremobates pulpitayulosus Solifugae
Nosopoga sp. Solifugae
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DNA105733 Pseudocellus sp. Guatemala
DNA105736 Pseudocellus sp. Mexico
DNA102708 Ricinoides atewa Ghana
DNA102720 Ricinoides aff. feae Senegal
DNA102692 Ricinoides feae Guinea Bassai
DNA102695 Ricinoides feae Guinea Bassai
DNA102716 Ricinoides feae Guinea Bassai
DNA102693 Ricinoides feae Guinea Bassai
DNA102694 Ricinoides feae Guinea Bassai
DNA102691 Ricinoides feae Guinea Bassai
DNA105886 Ricinoides megahanseni Ivory Coast
DNA105881 Ricinoides megahanseni Ivory Coast
DNA105885 Ricinoides megahanseni Ivory Coast
DNA105884 Ricinoides cf. afzelis Guinea
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DNA104741 Ricinoides cf. olomouca Cameroon
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DNA104744 Ricinoides cf. olomouca Cameroon
DNA102682 Ricinoides sp. Equatorial Guinea
DNA102688 Ricinoides sp. Equatorial Guinea
DNA102690 Ricinoides sp. Equatorial Guinea
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DNA104746 Ricinoides karschii Cameroon
DNA102686 Ricinoides cf. karschii Equatorial Guinea
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DNA104743 Ricinoides cf. karschii Gabon
DNA104747 Ricinoides cf. karschii Gabon

Figure 2. Maximum credibility tree for African Ricinulei and outgroup taxa, obtained by Bayesian inference in BEAST [19]. 95% Confidence intervals for ages are represented by bars at nodes. Clade posterior probabilities above 50% are indicated above nodes and maximum-likelihood bootstrap frequencies below nodes. Posterior distributions for nodes of interest are also depicted below the tree. Palaeogeographic maps from Ron Blakey, NAU Geology at http://jan.ucc.nau.edu/~rcb7/.
known fossil arachnid, a scorpion) and 445 Ma (oldest known fossil chelicerate, a horse-shoe crab). The Beast analysis was run for 60 million generations with sampling every 1000 generations.

3. Results and discussion

The results recover the monophyly of Ricinulei and its component genera, Cryptocellus, Pseudocellus and Ricinoides, with high support (figure 2). The origin of the group (the divergence from its sister group) is dated to around 250 Ma. The oldest known fossil ricinuleid (not considered part of the Neoricinulei crown group), dated to 319 Ma [24], is concordant with the 95% confidence interval on the tree, and independently corroborates the analytical results and biogeographic conclusions. Ricinoides is sister to one of the two Neotropical genera, Pseudocellus, suggesting that the entire diversification of Ricinulei predates the fragmentation of Gondwana. This biogeographic interpretation, previously proposed based on morphological evidence [25], is corroborated by the molecular dating. Based on the results presented here, the diversification of Ricinoides in Western and Central Africa occurred in the Late Cretaceous, around 88 ± 33 Ma. Specimens from Guinea and Ivory Coast group with those from Cameroon, Equatorial Guinea and Gabon, probably an artefact of missing data (lack of mitochondrial gene sequences for the former; table 1). Samples from the Ivory Coast group with the remaining West African samples in the maximum-likelihood analysis, as expected based on their similar morphology and geographical proximity. Among the West African samples, the divergence between Ricinoides atewa Naskrecki, 2008 and other species from Senegal and Guinea-Bissau is dated to around 44 Ma.

Tropical forest biodiversity is lower in Africa than in South America and Southeast Asia [26]. This difference has been attributed to extinctions caused by forest fragmentation and potentially even the complete disappearance of the forest during past periods of severe aridification [5]. This is exemplified by the low diversity of another ancient group of soil animals, velvet worms (Onychophora Grube, 1853), with a single species of Peripatidae Evans, 1901 in tropical Africa (Gabon and Cameroon), when compared with four species in Southeast Asia and 68 in the Neotropics [27]. The relatively low diversity of the tropical African forests is consistent with the majority of molecular evidence, which suggests that forested endemics are mostly recent (Late Miocene–Pleistocene) and diversified according to the refugial speciation model [4].

The results presented provide molecular evidence for an endemic African rainforest taxon, Ricinoides, the origin of which can be traced to the fragmentation of Gondwana, and confirms evidence from other ground-dwelling arthropods [28] for an early diversification in the Late Cretaceous, around 90 Ma, in Western and Central Africa. This period corresponds to a time of diversification among angiosperms and associated reduction in gymnosperm diversity, documented in the Cenomanian stage of the Late Cretaceous [29]. Even if the Mesozoic rainforests were structurally and compositionally different from those in present-day tropical Africa, stratified forests may have been present since the Late Cretaceous, based on the presence of large seeds and fruits in the fossil record [30]. The major divergence observed among species in Western and Central Africa around 90 Ma could be the result of vicariance caused by successive marine incursions (figure 2) that commenced in the Late Cretaceous [31]. Refugia may also have played a role in allopatric speciation on a smaller scale for more recent species in the area of present-day Cameroon and Gabon.

The results presented suggest further that ancient diversifications exist in Western and Central Africa and the biodiversity of this region may have been greater during Late Cretaceous to Palaeocene times. Climate change may have depleted diversity after the separation of Africa and South America, with subsequent stable refugia acting as ‘museums’ for ancient lineages. In this respect, the data are largely congruent with the fossil record, which suggests that entire lineages of Neotropical palms were present in Africa until at least the Late Oligocene (27–28 Ma) [7]. Such models [5] may have received little support from molecular studies until now (but see [6]), because few phylogeographic studies have been conducted on old lineages with high endemism and low population density (see electronic supplementary material). Furthermore, extinction rates are difficult to infer from molecular studies [32] in the absence of fossils from outside the putative refugia. Ricinoides thus represents one of the oldest endemic African genera for which the origin, early diversification and subsequent survival in Miocene forest refugia has been studied and tested phylogenetically.

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References

5. Hamilton AC. 1976 The significance of patterns of distribution shown by forest plants and animals in tropical Africa for the reconstruction of upper