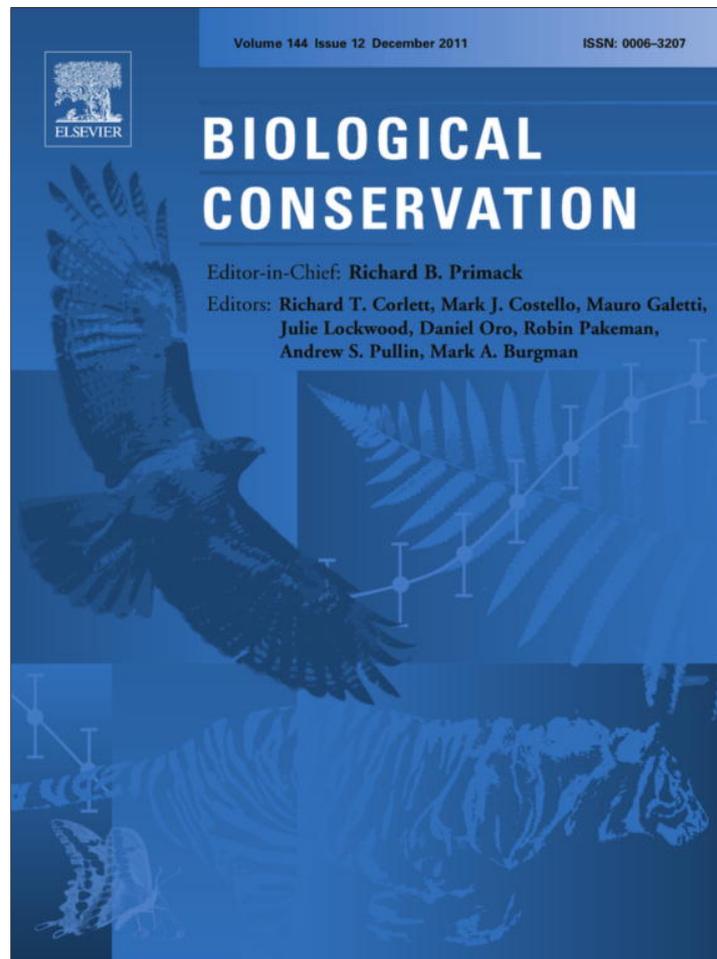


Provided for non-commercial research and education use.
Not for reproduction, distribution or commercial use.



This article appeared in a journal published by Elsevier. The attached copy is furnished to the author for internal non-commercial research and education use, including for instruction at the authors institution and sharing with colleagues.

Other uses, including reproduction and distribution, or selling or licensing copies, or posting to personal, institutional or third party websites are prohibited.

In most cases authors are permitted to post their version of the article (e.g. in Word or Tex form) to their personal website or institutional repository. Authors requiring further information regarding Elsevier's archiving and manuscript policies are encouraged to visit:

<http://www.elsevier.com/copyright>



Contents lists available at SciVerse ScienceDirect

Biological Conservation

journal homepage: www.elsevier.com/locate/biocon

Short communication

The impact of fossils on the Evolutionary Distinctiveness and conservation status of the Australian lungfish

Lionel Cavin^{a,*}, Anne Kemp^b^a Muséum d'histoire naturelle de la Ville de Genève, CP 6434, 1211 Genève 6, Switzerland^b Australian Rivers Institute, Griffith University, 170 Kessels Road, Nathan, Brisbane, Queensland 4111, Australia

ARTICLE INFO

Article history:

Received 4 July 2011

Received in revised form 24 August 2011

Accepted 28 August 2011

Available online 29 September 2011

Keywords:

Fossil record

Phylogeny

Evolutionary Distinctiveness index

Neoceratodus

Mesozoic

ABSTRACT

The recognition of phylogenetic information for evaluating conservation priorities has stressed the importance of basal taxa. The “Evolutionary Distinctiveness” index (ED) is a species-specific index that includes branch length expressed as an absolute value measured in millions of years that can be applied to a single terminal taxon in a phylogeny. The ED depends on the tree pattern, i.e. of a cladogram included into a time-scale. When calculated for the Australian lungfish (*Neoceratodus forsteri*), a threatened dipnoan that occurs naturally only in southeast Queensland, the ED index shows variable value according to the chosen tree. On the basis of a recently proposed phylogeny including a new fossil find from the Early Cretaceous of Thailand, the ED value reaches the highest value for piscine sarcopterygians, and for all vertebrates, and thus reinforces the “originality” of this fish. This example points out the importance of fossils in the resolution of phylogenies and beyond, in the calculation of indexes supporting conservation decisions.

© 2011 Elsevier Ltd. All rights reserved.

1. Introduction

The Australian lungfish (*Neoceratodus forsteri*) occurs naturally only in the Brisbane, Burnett and Mary rivers in southeast Queensland. Despite low genetic diversity (Frentiu et al., 2001), sufficient information has emerged from DNA fingerprinting studies to show that Brisbane River fish are distinct from Mary River fish (Lissone, 2003). The Australian lungfish was once widespread in Queensland (Kemp, 1987), and a small population was even found in the Murray River in Victoria in the Pliocene (Kemp, unpublished; collection of the Museum of Victoria, Melbourne). The species may now be restricted to a few rivers and lakes in southeast Queensland, but it is still true to say that they have weathered many natural disasters in the past, including periodic droughts that may last for several years, and severe flooding that destroys habitat, carries fish well away from their preferred homes, and removes food supplies. Since white settlement in southeast Queensland, these problems have been accentuated. Most of the difficulties faced by living lungfish arise, not from natural disasters alone, but from human interference with their environments, making any natural disasters much worse. Population pressure in southeast Queensland, and the demands of agriculture and industry for water (Arthington, 2009), has resulted in massive environmental degradation in the three rivers where

* Corresponding author.

E-mail addresses: Lionel.cavin@ville-ge.ch (L. Cavin), a.kemp@griffith.edu.au (A. Kemp).

lungfish occur naturally (Kemp, 1987, 1995), and other systems such as the Coomera and Pine Rivers, where lungfish have been introduced (O'Connor, 1896; Welsby, 1905). The main threats that affect lungfishes are droughts, transformation of the aquatic vegetation affecting the courtship ritual, egg deposition and availability of suitable habitats for the young fishes (Kemp, 2005, in press), restriction of migratory behavior (Kind, 2011) caused by the presence of dams, and the development of bacteria and fungi on the eggs (Kemp, 1994, in press). Inadequate nutrition of adult lungfish in water impoundments created by reservoirs has resulted in poor development of eggs and young produced by these adults (Kemp, in press).

The Australian lungfish is listed in Appendix II of the Convention on International Trade in Endangered Species (CITES). The Australian lungfish is also classified as vulnerable to extinction (Environment Protection and Biodiversity Conservation Act, 1999) and protected by several Queensland State Government laws, in 1914, 1972 and 1994. Queensland State Government officials insist that the lungfish are common within their range, although in fact they may only be locally abundant in certain places. Despite the laws protecting lungfish, the species remains at risk of extinction, as has already happened in Enoggera Reservoir, and may occur in other water impoundments if steps are not taken to permit successful spawning and recruitment of young to the adult population (Kemp, in press). State Government officials in Queensland have designed a recovery plan for lungfish affected by changes to their environments, although this has not

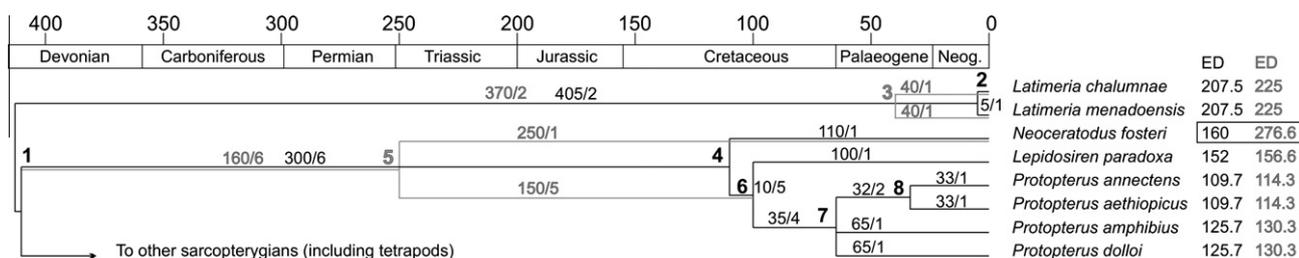


Fig. 1. Phylogeny of living piscine sarcopterygians in a time scale (tree). Fraction numbers above branches correspond to the length of the branch in millions years divided by the number of its descendent species. ED, or Evolutionary distinctness, correspond to the sum of the values for branches from which species is descended, to the root of the phylogeny (here the common ancestor of sarcopterygians). Gray branches and values correspond to alternative trees. Bold numbers indicates divergence events or oldest occurrences. (1) Oldest dipnoan remains, Early Devonian, Pragian, ca. 410 myrs, according to Clément et al. (2006). (2) Divergence between *L. chalumnae* and *L. menadoensis*, 5 myrs ago, according to Holder et al. (1999) on the basis of partial mitochondrial gene sequences. (3) Divergence between *L. chalumnae* and *L. menadoensis*, 40 myrs ago, according to Inoue et al. (2005) on the basis of the whole mitochondrial genome sequence. (4) divergence between *Neoceratodus* and Lepidosireniformes in the Early Cretaceous, Albian, 110 myrs, on the basis of the oldest fossil record of *Neoceratodus* (Kemp and Molnar, 1981) and on a *Neoceratodus*/Lepidosireniformes sister relationships (for instance Schultze, 2004). (5) Divergence between *Neoceratodus* and Lepidosireniformes in the Early Triassic, 250 myrs, on the basis of the phylogeny of Cavin et al. (2007), with recognition of stem Lepidosireniformes in the Early Triassic (not shown on the figure). (6) Divergence between the *Lepidosiren* and *Protopterus* lineages, 100 myrs, based on the analysis of the African fossil record (Otero, 2011). (7) Basal divergence of the extant *Protopterus* species, Cretaceous-Tertiary transition, ca 65 myrs, according to Tokita et al. (2005) on the basis of partial sequences of the mitochondrial 16S rRNA. (8) Divergence between *P. annectens* and *P. aethiopicus* in “mid-Tertiary”, ca. 33 myrs, according to Tokita et al. (2005).

yet been effective in improving the conditions of lungfish trapped in water impoundments, or in rivers affected by floods and droughts.

The Australian lungfish lives for many years, and low recruitment of juveniles to the population means that habitats never have too many lungfish (Kemp, 1995). However, young lungfish are vulnerable, and need the protection of a natural habitat, where the adult lungfish are able to carry out their courtship rituals, and deposit the eggs on the leaves and rootlets of water plants (Kemp, 1987). Few of the eggs laid under these circumstances are unfertilised, and most of the eggs that survive predation develop to hatching (Kemp 1994, in press). In reservoirs and lakes, with-out submerged plants, adults shed the eggs loose into the water. Many are not fertilised, and there are no places for young lungfish to hide. This leaves young lungfish exposed to predation, and there is little food. Certainly it is true that, apart from exceptional years (Kemp, 1987), juvenile lungfish are rarely found, and catches consist of large adult fish only. When recruitment was prevented in Enoggera Reservoir after juvenile habitats were destroyed in 1974, the lungfish population slowly became extinct (Kemp, 2005, in press). This will happen eventually in the large artificial lakes created by dams and weirs, if eggs and young have no refuges, and adult lungfish cannot find suitable food and any eggs that they produce are deficient and fail to develop (Kemp, in press).

2. Methods

Neoceratodus is the most primitive living member of the Dipnoan lineage (Dipnoi). The recognition of phylogenetic information for evaluating conservation priorities has stressed the importance of basal taxa (Stiassny and de Pinna, 1994), and policies for biological conservation now integrate the “originality” (sensu Pavoine et al., 2005) of species. Quantitative approaches have been proposed to provide objective grounds supporting conservation decisions (for instance Mooers et al., 2008). But these methods have rarely been applied, if ever, to the Australian lungfish. Several existing indexes are based on tree topology, but do not consider branch length (Vane-Wright et al., 1991; May, 1990). Other methods include branch length in their calculation, such as the “phylogenetic diversity” (PD) index introduced by Faith (1992): it corresponds to the sum of the length of all the branches in a phylogenetic tree. Branch length in a cladogram corresponds to the number of the steps (transformation of characters) along the branch, which may correspond either to molecular or to morpho-

logical features. Branch length is not proportional to time duration because it depends of the evolutionary rate of the lineage under study. Lungfishes have a slower rate of evolution of their genome as compared to tetrapods (Lee et al., 2006), and a low rate of morphological evolution in the last part of their evolutionary history, i.e. after the Carboniferous (Schultze, 2004). In order to better integrate these evolutionary characteristics and to include data from the fossil record, we favor here methods that integrate time as an absolute value. Altschul and Lipman (1990) showed the importance of branch length when comparing species in the perspective of conservation biology. Redding et al. (2008) expressed this view mathematically, the pendant edge (PE), which corresponds to an absolute value measured in millions of years. These authors showed that the PE captures well the phylogenetic diversity (PD) of a group. Isaac et al. (2007) defined a species-specific index, the “Evolutionary Distinctiveness” index (ED), rather similar to the PE value. This method is used here because it can be applied to a single terminal taxon in a phylogeny (on the contrary, PD deals with a set of taxa). The method is detailed in the caption of Fig. 1.

3. Results

We calculated the “Evolutionary Distinctiveness” (ED) for *N. forsteri*, as well as for the other living piscine sarcopterygians. There is a consensus about the phylogeny of living lungfishes: the African genus *Protopterus* is the sister genus of the South American *Lepidosiren*, both forming the sister group of the Australian lungfish. When fossil taxa are included in the analysis, most cladograms also show the pattern ((*Protopterus*–*Lepidosiren*) – *Neoceratodus*) with the fossil taxa resolved as stem Dipnoi (Martin, 1982; Marshall, 1987; Thomson, 1990; Schultze and Chorn, 1997; Schultze, 2004). On the basis of this pattern, the fossil record of the recent genera indicates a dichotomy between *Lepidosiren* and *Protopterus* at ca. 100 myrs, and a dichotomy between that lineage and the *Neoceratodus* lineage at ca. 110 myrs (Fig. 1). However, an alternative phylogenetic analysis including new Mesozoic taxa, notably *Ferganoceratodus martini* known by a skull roof described from the Early Cretaceous of Thailand, and resting on new definitions of characters led to an alternative hypothesis of relationships (Cavin et al., 2007). In this pattern, several Triassic and Jurassic lineages are resolved as stem Lepidosireniformes (the order that gathers Lepidosirenidae + Protopteridae). The dichotomy between *Neoceratodus* and all other living lungfishes should be as old as the oldest stem Lepidosireniformes, i.e. Early Triassic in age, ca. 250 myrs.

Fig. 1 shows both patterns and their implications on the calculation of the ED indexes. In the first phylogenetic hypothesis, the ED index of *N. forsteri* equals 160. This is slightly superior to ED index of *Lepidosiren paradoxa* (152) and to *Protopterus* spp. (125.7 and 109.7). This index is lower than those calculated for both species of *Latimeria*, whatever age is retained for the separation of both living species (ED = 207.5 according to a divergence time of 5 myrs (Holder et al., 1999) and ED = 225 according to a divergence time of 40 myrs (Inoue et al., 2005)). In the second hypothesis, i.e. an individualization of the *Neoceratodus* lineage 250 myrs ago, the ED of that species rises to 276.6. This makes it significantly higher than the ED of *Lepidosiren* (156.6) and *Protopterus* spp. (130.5 and 114.4), and also higher than the ED index of *Latimeria* spp. (225). Mention should be made that with a single living species of coelacanth, i.e. the situation before the description of *L. menadoensis* in 1999, the ED index of *L. chalumnae* would have been of 410).

4. Discussion

Is not possible to compare the ED value of *Neoceratodus* to published values for other sarcopterygians (for instance values for mammals (Isaac et al., 2007)), because those are calculated from the common ancestor of the group under study (mammals) and not from the common ancestor of all sarcopterygians. However, we can safely hypothesize that the ED value for *N. forsteri* is the highest among sarcopterygians, and among vertebrates, because (1) only two nodes separates the terminal taxon from the common ancestor of sarcopterygians (one dichotomy with tetrapods and one dichotomy with other lungfishes) and (2) because of the very long terminal branch (120 or 250 myrs according to the chosen phylogeny). All other putative vertebrate candidates, usually regarded as living fossils, show either shorter terminal branches (for instance the bichirs, reedfishes, gars) or have much more nodes separating them from the common ancestor of vertebrates (for instance the armadillo, tuatara, bowfin).

5. Conclusion

Isolated tooth plates of *N. forsteri* were found in the Early Cretaceous (Albian) of Lightning Ridge, Griman Creek Formation, New South Wales (Kemp and Molnar, 1981). This occurrence makes this species (and this genus) the oldest surviving species of vertebrate. The new data presented in this paper indicates, moreover, that *N. forsteri* display the highest ED index among vertebrates, and points out the uniqueness of this fish and the importance of its conservation. Beyond the impact of fossils on calculation of indexes supporting (or not) conservation decisions, we notice that a better understanding of the fossil record not only unveils the contingent and unique history of life on Earth, but also impacts on the “evolutionary significance” of living organisms and consequently on their conservation status.

Acknowledgments

L.C. was supported by a grant from the Swiss National Science Foundation (200021-113980) and A.K. was supported by the Adelphi Australia Science Foundation. We thank the three anonymous referees for their valuable comments and corrections.

References

- Altschul, S.F., Lipman, D.J., 1990. Equal animals. *Nature* 348, 493–494.
 Arthington, A.H., 2009. Australian lungfish, *Neoceratodus forsteri*, threatened by a new dam. *Environmental Biology of Fishes* 84, 211–221.

- Cavin, L., Suteethorn, V., Buffetaut, E., Tong, H., 2007. A new Thai Mesozoic lungfish (Sarcopterygii, Dipnoi) with an insight into post-Palaeozoic dipnoan evolution. *Zoological Journal of the Linnean Society* 149, 141–177.
 Clément, G., Dupret, V., Goujet, D., Pernègre, V., Roy, J.-C., 2006. First Devonian dipnoans (Vertebrata, Sarcopterygii) from Spitsbergen. *Comptes Rendus Palevol* 5, 890–893.
 Faith, D.P., 1992. Conservation evaluation and phylogenetic diversity. *Biological Conservation* 61, 1–10.
 Frentiu, F.D., Ovenden, J.R., Street, R., 2001. Australian lungfish (*Neoceratodus forsteri*: Dipnoi) have low genetic variation at allozyme and mitochondrial DNA loci: a conservation alert? *Conservation Genetics* 2, 63–67.
 Holder, M.T., Erdmann, M.V., Wilcox, T.P., Caldwell, R.L., Hillis, D.M., 1999. Two living species of coelacanths? Proceedings of the National Academy of Sciences of the USA 96, 12616–12620.
 Inoue, J.G., Miya, M., Venkatesh, B., Nishida, M., 2005. The mitochondrial genome of Indonesian coelacanth *Latimeria menadoensis* (Sarcopterygii: Coelacanthiformes) and divergence time estimation between the two coelacanths. *Gene* 349, 227–235.
 Isaac, N.J.B., Turvey, S.T., Collen, B., Waterman, C., Baillie, J.E.M., 2007. Mammals on the EDGE: conservation priorities based on threat and phylogeny. *PLoS ONE* 2, e296.
 Kemp, A., 1987. The biology of the Australian lungfish, *Neoceratodus forsteri*. *Journal of Morphology* (Suppl. 1), 181–198.
 Kemp, A., 1994. Pathology in eggs and embryos of *Neoceratodus forsteri*. (Osteichthyes: Dipnoi). *Copeia* 1994, 435–443.
 Kemp, A., 1995. Threatened fishes of the world: *Neoceratodus forsteri* (Kreffft 1870) (Neoceratodontidae). *Environmental Biology of Fishes* 43, 310.
 Kemp, A., 2005. New insights into ancient environments using dental characters in Australian Cretaceous lungfish. *Alcheringa* 29, 123–149.
 Kemp, A., in press. Comparison of embryological development in the threatened Australian lungfish, *Neoceratodus forsteri*, from two sites in a Queensland river system. *Endangered Species Research*.
 Kemp, A., Molnar, R.E., 1981. *Neoceratodus forsteri* from the Lower Cretaceous of New South Wales, Australia. *Journal of Paleontology* 55, 211–217.
 Kind, P.K., 2011. The natural history of the Australian Lungfish *Neoceratodus forsteri* (Kreffft, 1870). In: Jørgensen, J.M., Joss, J. (Eds.), *Biology of Lungfishes*. CRC Press, Enfield, New Hampshire, pp. 61–96.
 Lee, J., Alrubaian, J., Dores, R.M., 2006. Are lungfish living fossils? Observation on the evolution of the opioid/orphanin gene family. *General and Comparative Endocrinology* 148, 306–314.
 Lissonne, I., 2003. Conservation Genetics and the Australian Lungfish *Neoceratodus forsteri* (Kreffft 1870): A Spatio-temporal Study of Population Structure. Master of Science Thesis, Faculty of Science, The University of the Sunshine Coast, Sippy Downs, Queensland.
 Marshall, C.R., 1987. Lungfish: phylogeny and parsimony. *Journal of Morphology* (Suppl. 1), 151–162.
 Martin, M., 1982. Nouvelles données sur la phylogénie et la systématique des dipnoi postpaléozoïques. *Comptes Rendus de l'Académie des Sciences, série II* 294, 611–614.
 May, R.M., 1990. Taxonomy as destiny. *Nature* 347, 129–130.
 Mooers, A.Ø., Faith, D.P., Maddison, W.P., 2008. Converting endangered species categories to probabilities of extinction for phylogenetic conservation prioritization. *PLoS ONE* 3, e3700.
 O'Connor, D., 1896. On the redistribution of *Ceratodus*. In: *Proceedings of the Royal Society of Queensland* 11, pp. ii and xvi.
 Otero, O., 2011. Current knowledge and new assumptions on the evolutionary history of the African lungfish, *Protopterus*, based on a review of its fossil record. *Fish & Fisheries* 12 (3), 235–255.
 Pavoine, S., Ollier, S., Dufour, A.-B., 2005. Is the originality of a species measurable? *Ecology Letters* 8, 579–586.
 Redding, D.W., Hartmann, K., Mimoto, A., Bokal, D., DeVos, M., Mooers, A.Ø., 2008. Evolutionarily distinctive species often capture more phylogenetic diversity than expected. *Journal of Theoretical Biology* 251, 606–615.
 Schultze, H.-P., 2004. Mesozoic sarcopterygians. In: Arratia, G., Tintori, A. (Eds.), *Mesozoic Fishes 3 – Systematics, Palaeoenvironments and Biodiversity*. Verlag Dr Friedrich Pfeil, München, pp. 463–492.
 Schultze, H.-P., Chorn, J., 1997. The Permo-Carboniferous genus *Sagenodus* and the beginning of modern lungfish. *Contributions to Zoology* 67, 9–70.
 Stiassny, M.L.J., De Pinna, M.C.C., 1994. Basal taxa and the role of cladistic patterns in the evaluation of conservation priorities: a view from freshwater. In: Forey, P.L., Humphries, C.J., Vane-Wright, R.I. (Eds.), *Systematics and Conservation Evaluation*. Systematics Association Special, vol. 50. Clarendon Press, Oxford, pp. 235–249.
 Thomson, K.S., 1990. Triassic Fishes from the Cassange Depression (R.P. de Angola). III.2.3-Sarcopterygii/Dipnoi. *Ciencias da Terra (Numero Especial)*, pp. 29–33.
 Tokita, M., Okamoto, T., Hikida, T., 2005. Evolutionary history of African lungfish: a hypothesis from molecular phylogeny. *Molecular Phylogenetics and Evolution* 35, 281–286.
 Vane-Wright, R.I., Humphries, C.J., Williams, P.H., 1991. What to protect systematics and the agony of choice. *Biological Conservation* 55, 235–254.
 Welsby, T., 1905. *Schnappering and Fishing in the Brisbane River and Moreton Bay Waters, A Wandering Discourse on Fishing Generally*. Outridge Printing Co., Brisbane.