

Research



Cite this article: Barrett PM, Evans DC, Campione NE. 2015 Evolution of dinosaur epidermal structures. *Biol. Lett.* **11**: 20150229. <http://dx.doi.org/10.1098/rsbl.2015.0229>

Received: 23 March 2015

Accepted: 7 May 2015

Subject Areas:

palaeontology, evolution

Keywords:

Dinosauria, integument, scales, feathers, phylogeny

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Electronic supplementary material is available at <http://dx.doi.org/10.1098/rsbl.2015.0229> or via <http://rsbl.royalsocietypublishing.org>.

Evolution of dinosaur epidermal structures

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Spectacularly preserved non-avian dinosaurs with integumentary filaments/feathers have revolutionized dinosaur studies and fostered the suggestion that the dinosaur common ancestor possessed complex integumentary structures homologous to feathers. This hypothesis has major implications for interpreting dinosaur biology, but has not been tested rigorously. Using a comprehensive database of dinosaur skin traces, we apply maximum-likelihood methods to reconstruct the phylogenetic distribution of epidermal structures and interpret their evolutionary history. Most of these analyses find no compelling evidence for the appearance of protofeathers in the dinosaur common ancestor and scales are usually recovered as the plesiomorphic state, but results are sensitive to the outgroup condition in pterosaurs. Rare occurrences of ornithischian filamentous integument might represent independent acquisitions of novel epidermal structures that are not homologous with theropod feathers.

1. Introduction

Feathered and protofeathered non-avian theropods provide critical evidence in debates over bird origins and dinosaur biology [1]. These include specimens with soft-tissue preservation from the Late Jurassic–Late Cretaceous of Germany [2], Canada [3] and China [4,5]. Quill-like, protofeather-like and branched structures have been recorded in ornithischians [6–8] and this, combined with their ubiquitous occurrence among coelurosaurian theropods, has nurtured suggestions that dinosaurs primitively possessed a complex integumentary covering in addition to the epidermal scales present in many taxa [2,9,10].

Inferred possession of protofeathers in the dinosaur common ancestor has profound implications for understanding their biology and evolution, especially locomotion, physiology and behaviour [1,11,12]. However, this hypothesis remains untested within the combined context of the fossil record of dinosaur skin and an explicit phylogenetic framework. Here, we test the hypothesis that dinosaurs were plesiomorphically protofeathered by estimating the phylogenetic distribution of dinosaur integumentary structures. We also discuss preservational biases and their impact on the recovered pattern.

2. Material and methods

We constructed a comprehensive database of non-avian dinosaur integumentary occurrences based on an existing compilation [13], primary literature sources (see the electronic supplementary material, S1) and personal observations (P. M. Barrett and D. C. Evans 1996–2015). The database includes 34 ornithischian, six sauropod and 40 theropod taxa (including several Mesozoic birds), plus several generically indeterminate records (electronic supplementary material, S1). All taxa were scored for the presence/absence of epidermal scales, unbranched filaments (protofeathers)/quills and more complex branched filaments (including feathers). Taxa with ulnar quill

knobs were assumed to possess complex branched filaments/feathers [14]. The body regions preserving these integumentary features (e.g. forelimb and tail) were recorded to assess the completeness of the integumentary record and facilitate comparisons between taxa (e.g. if filaments were primarily present on tails, but not elsewhere; electronic supplementary material, S1). Recent phylogenies were used to construct an informal consensus tree containing all taxa with known epidermal structures (electronic supplementary material, S2). The sedimentary environments in which skin-bearing specimens were preserved were recorded to investigate the role of taphonomy in the phylogenetic distribution of integumentary structures.

Ancestral distributions of integumentary structures were analysed using maximum-likelihood approaches. Maximum-likelihood reconstructions of ancestral character states for discrete characters were conducted using the 'ace' function in the ape package [15], with tree branch lengths estimated in terms of time, derived using the 'timePaleoPhy' function in the paleotree package [16], all implemented in R. Branch lengths were estimated using two time-scaling methods in order to test the sensitivity of the pattern to scaling method [17]: 'minimum branch length' (mbl), which minimizes inferred branching times and closely resembles the raw, time-calibrated tree, and 'equal branch length' (equal), which adds a pre-designated length to the tree root (1 Ma) and then evenly distributes zero-length branches. The latter technique generally results in more evenly distributed branch lengths throughout the tree but tends to extend terminal branching events back in time, especially when internal ghost lineages are extensive. Accordingly, terminal taxa have a greater effect on the root node under these conditions. A third set of analyses was run with all branch lengths scaled to unity, which standardizes the effect of surround taxa and nodes on ancestral likelihoods. Maximum-likelihood analyses were run using binary/multistate and ordered/unordered characterizations of integumentary characters. Skin impressions are unknown for non-dinosaurian dinosauromorphs, so pterosaurs were selected as the outgroup. Some pterosaurs possessed filamentous coats [18], but it is unknown if this is derived/primitive for the clade: consequently, each of the aforementioned analyses was conducted with pterosaurs scored as either primitively scale- or filament-covered, with the assumption that filaments were homologous to dinosaur filaments. An ANOVA was conducted on the results of the maximum-likelihood analyses to determine the influence of these different parameters on nodal likelihood values (electronic supplementary material, S2).

The null hypotheses that different preservational environments (alluvial versus lacustrine/lagoonal) or individual body regions (e.g. tail and forelimb) preserved epidermal scales or filamentous integument with equal probability were tested with χ^2 analyses (electronic supplementary material, S2).

3. Results

Regardless of branch-scaling method or character construction (binary/multistate, ordered/unordered), analyses assuming a scaled pterosaur ancestor recovered Dinosauria and Ornithischia as primitively scaled with scaled likelihoods ranging from 0.92–1.00 and 0.79–0.97, respectively (figure 1a: electronic supplementary material, S1 and S2). Analyses assuming a filament-covered pterosaur ancestor produced different results, with a minority recovering Dinosauria and Ornithischia as primitively filament-covered with lower scaled likelihoods ranging from 0.05–0.82 and 0.19–0.85, respectively (figure 1b: electronic supplementary material, S1 and S2). Twelve of the 18 analyses recovered nodal likelihood values of more than 0.50 in support of

scaled ancestors for both Dinosauria and Ornithischia, with most of these recovering likelihood values of more than 0.80 (figure 1c: electronic supplementary material, S1 and S2). Support for a filament-covered saurischian or theropod ancestor varied between analyses, but the coelurosaur ancestor was recovered frequently as primitively filament-covered with likelihood values ranging from 0.15–1.0 and 15 of 18 analyses finding support of more than 0.50 for this scenario (figure 1c: electronic supplementary material, S1 and S2). An ANOVA indicates that the epidermal covering of the pterosaur outgroup has the strongest effect on the nodal likelihood values recovered by these analyses, whereas branch-scaling methods and character scoring are less important (electronic supplementary material, S2).

A χ^2 test indicates that the type of epidermal structure preserved is strongly associated with depositional environment. Filaments/feathers are more frequently preserved in lacustrine/lagoonal settings than would be expected by chance ($p < 0.0001$; electronic supplementary material, S2). Conversely, there is no significant correlation between the body region preserved and the integumentary structure type present ($p = 0.1239$; electronic supplementary material, S2).

4. Discussion

Current data do not provide definitive support for the suggestion that protofeathers were synapomorphic for dinosaurs. Instead, it seems most likely that scaly skin, unadorned by feathers or their precursors, was primitive for Dinosauria and retained in the majority of ornithischians, all sauropodomorphs and some early-diverging theropods (filaments are thus far unknown in ceratosaurians, abelisaurids and allosauroids: electronic supplementary material, S1). Additional examples of protofeathers would be required from early dinosaur lineages or non-dinosaurian dinosauromorphs to optimize this feature to the base of Dinosauria. In particular, the ancestral condition in pterosaurs is pivotal in this regard, but currently unknown. Filaments/feathers were probably present in the coelurosaur common ancestor and might have characterized all theropods, though support for the latter proposal is equivocal (electronic supplementary material, S1 and S2). There is some evidence across the dinosaur tree for homoplastic loss of these structures (e.g. some tyrannosaurids possess scales, but lack evidence for other epidermal structures; electronic supplementary material, S1), so integument evolution cannot be regarded as a progression to more complex epidermal structures.

Caution is warranted as taphonomy controls the types of integumentary structures preserved in different taxa. Feather-like structures are most frequently preserved in low-energy environments and may have been lost in coarse-grained, high-energy settings, though rare feather-like structures are known from the latter [3]. No dinosaur skin impressions (except footprints) are known from the Late Triassic and they are rare from the Early–Middle Jurassic (electronic supplementary material, S1) periods when dinosaur-bearing lacustrine/lagoonal deposits are scarce. Hence, potential taphonomic windows for early dinosaur filaments/feathers are not available currently: new localities are needed to resolve whether early dinosaurs were scaled or feathered. Conversely, the exceptional preservation of coelurosaurs in Late Jurassic and Cretaceous lagerstätte might not be indicative of skin structures in earlier relatives.

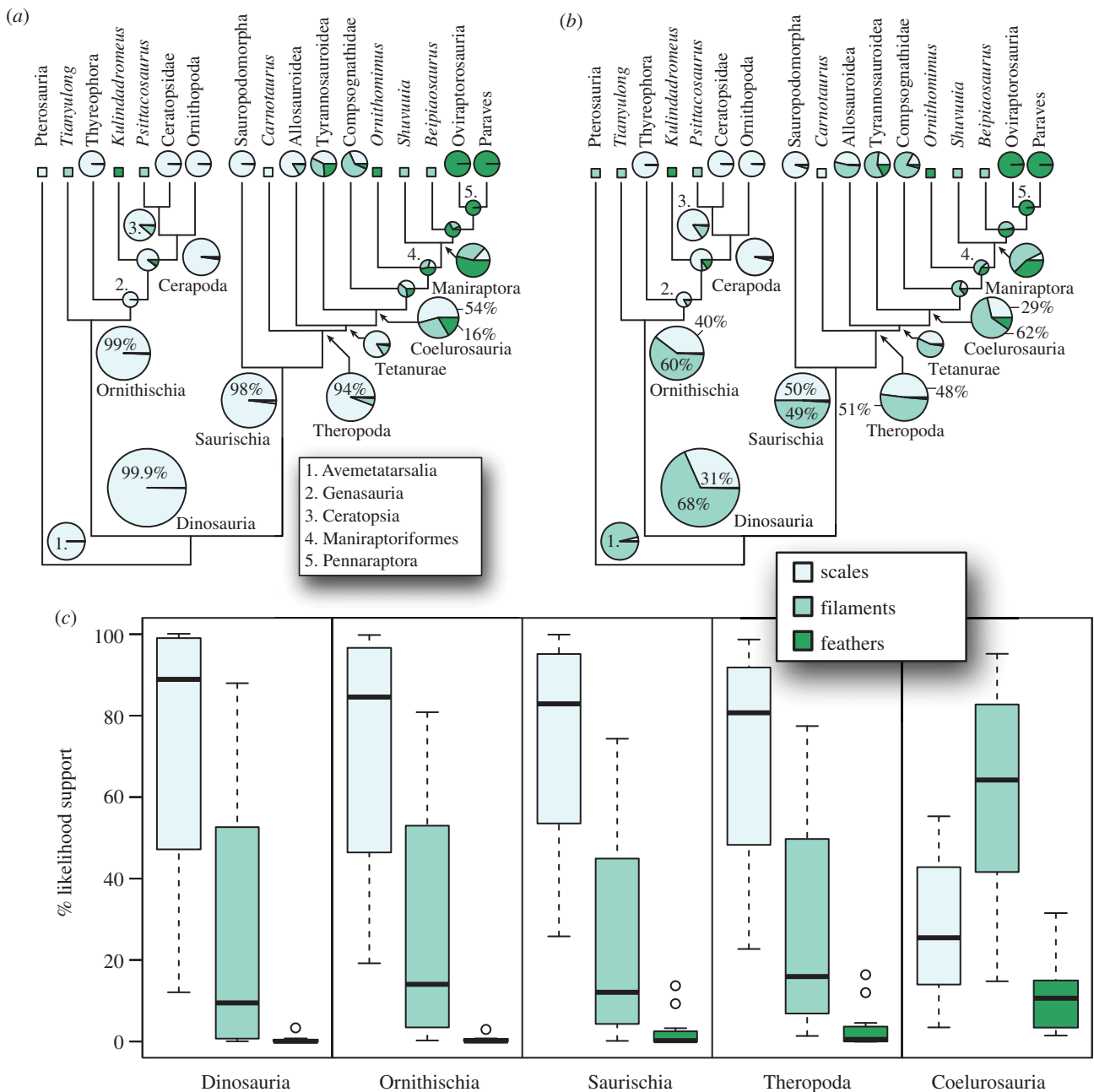


Figure 1. Ancestral state reconstructions for ornithodiran integument. Ancestral states based on unordered character states across the 'equal' time-scaled tree (*a–b*): pterosaurs coded as (*a*) plesiomorphically scaled and (*b*) filament-covered. (*c*) Range of probabilities for each state at particular nodes based on all permutations described in the text. Percentages reflect the relative support for a given state with respect to the others. (Online version in colour.)

Previous authors noted that ornithischian quills/ protofeathers were morphologically distinct from those in theropods [6,19]. Our analyses support suggestions that these features should not be regarded *a priori* as homologous with theropod epidermal structures: ornithischian quills/ protofeathers plausibly represent epidermal structures that evolved independently, and may be indicative of a more general ornithodiran tendency to experiment with epidermal features. The latter possibility is suggested by: possession of feathers and protofeathers in theropods [2–5]; presence of filamentous coverings in at least some pterosaurs [18]; and development of elaborate midline scale frills in hadrosaurs and sauropods (electronic supplementary material, S1). As archosaur scales, claws and feathers are composed of β -keratins [20,21], it is possible that the elaboration of all complex ornithodiran epidermal structures was underpinned by the same developmental and regulatory mechanisms ([22]:

which, for unknown reasons, were not expressed in the majority of non-coelurosaurian dinosaurs). Molecular phylogenies of β -keratin families indicate that those found in feathers are the latest diverging among archosaurs and may not have appeared prior to the evolution of crown birds, whereas scale and 'feather-like' β -keratins diverged earlier [21]. This suggests a scenario in which scales and feather-like structures may have appeared (and diversified) via numerous independent acquisitions in Ornithodira, with true feathers appearing only in birds and their proximate theropod outgroups.

The identification of branched feathers in theropods is uncontroversial and supported by morphology, developmental models and known β -keratin composition in some taxa [23]. Phylogenetic congruence and developmental data also support the homology of branched feathers and theropod filamentous protofeathers [23]. However, gross morphological

differences are not sufficient to establish homologies between true feathers and ‘protofeathers’ in other ornithodiran lineages as these structures potentially represent modified epidermal scales (like those of *Iguana*, *Kolekanos* or *Longisquama* [6,24]) or other tissues (e.g. degraded collagen fibres [25]). Additional homology tests are required, such as identification of β -keratin antibody reactivity, detailed comparisons between collagenous and epidermal structures, and identification of microstructural differences between elongate scales and protofeathers [5,22,26]. However, as reptilian scale and feather β -keratins are compositionally similar [20,21], it may be difficult to distinguish elongate scales from genuine protofeathers on the basis of biogeochemistry and gross morphology.

Current data indicate that feathers and their filamentous homologues are probably theropod synapomorphies but fail

to support the hypothesis that protofeathers are plesiomorphic for Dinosauria. The origins of ‘protofeathers’ and ‘quills’ outside Theropoda are ambiguous: interpretations of their homology should be correspondingly cautious, especially in those ornithodirans phylogenetically distant from birds.

Data accessibility. Supporting data are in the electronic supplementary material.

Authors’ contributions. P.M.B. and D.C.E. collected the data; N.E.C. and P.M.B. analysed the data; P.M.B. wrote the manuscript with assistance from D.C.E. and N.E.C.

Competing interests. We declare we have no competing interests.

Funding. D.C.E. is funded by NSERC (Canada).

Acknowledgements. We thank R. Benson, R. Butler, M. Davis and four referees for comments.

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