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The phylogenetic significance of bone types in euteleost fishes

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The paradentary is a small, sometimes dentigerous element in the lower jaw of some atherinomorph neoteleost fishes. Identification of the paradentary as a neomorphic, perichondrally ossified bone prompted re-examination of theories of the association of bone and teeth in teleost fishes. Teeth on a chondral lower jaw bone might be explained simply by epidermal–mesodermal interactions. Since the work of Kölliker in 1859, it has been known that there are two basic types of bone in teleost fishes: cellular bone, characterized by a matrix that has enclosed osteoblasts or osteocytes; and acellular bone, characterized by a relatively featureless matrix that lacks these bone-forming cells. Cellular bone is typical of lower teleosts, whereas acellular bone is typical of higher teleosts. Ontogenetic data indicate that acellular bone is derived relative to cellular bone. Even though identification of cellular and acellular bone can be made readily with histological preparations, acellular bone has been used infrequently as a character in analyses of teleost phylogeny. Acellular bone is considered here to be a derived character within teleost fishes. It is found in all Neoteleostei as well as some, but not all Salmoniformes. Independent of studies of bone, derived types of teeth in teleosts have been described in terms of their failure to become completely mineralized. Acellular bone and teeth of higher teleosts share several properties, including a large fraction of collagen. Teleosts lack a parathyroid gland; bone type is critical to the mechanism of calcium regulation. It is proposed that the character of acellular bone be incorporated into phylogenetic analyses of teleost fishes by correlating it with derived types of tooth structure.

KEY WORDS:—Acellular bone – cellular bone – chondral bone – dermal bone – teeth – calcium regulation – epidermal – mesodermal interactions – Euteleostei – Salmoniformes.

CONTENTS

INTRODUCTION

Since the work of Kölliker (1859a, b) it has been known that there are two major types of bone in teleosts, cellular and acellular. Cellular bone, like that of

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tetrapods, is characterized by enclosed osteoblasts or osteocytes in the bone matrix. Acellular bone lacks these bone-forming cells; the matrix is relatively featureless (Moss, 1961a, b, 1965).

There are degrees of cellularity, hence acellularity, among major groups of teleosts (Moss, 1961b, and references therein). However, these divisions are not always recognized and it is simplest and most useful for the purpose of this discussion to classify teleost bone as either cellular or acellular (as in Bertin, 1958; Ørvig, 1951; Moss, 1961a, b, 1965).

It was clear from Kölliker's work that teleosts could be divided into so-called lower teleosts and higher teleosts on the basis of bone type. Lower teleosts, such as osteoglossomorphs, elopomorphs and clupeomorphs (using the classification of Greenwood *et al.*, 1966), have cellular bone, whereas higher teleosts, such as percomorphs, atherinomorphs and paracanthopyterygians (using the classification of Rosen, 1973), have acellular bone.

Berg (1940) used cellular and acellular bone as characters in his classification of teleost fishes. Usually citing Kölliker, Berg stated whether a particular group of fishes had cells in the bone matrix or not. Despite the apparent phylogenetic significance of acellular bone, the character is mentioned rarely, if at all, in subsequent teleost classifications, such as those of Fink & Weitzman (1982), Greenwood *et al.* (1966), Lauder & Liem (1983), G. Nelson (1969), J. Nelson (1976, 1984) and Rosen (1973). This omission invites explanation. One possible reason is that acellular bone is known in primitive fossil vertebrates such as pteraspidomorph agnathans (e.g. see Ørvig, 1951; Stensiö, 1958, Schaeffer, 1977) and in the dermal skeleton and endoskeletal calcifications of some living and fossil chondrichthyans (Schaeffer, 1977; Peignoux-Deville & Janvier, 1984). Therefore, acellular bone has been termed an 'ancient tissue' (Moss & Posner, 1961) and is thought to be phylogenetically primitive. However, as discussed below, acellular bone is considered here to be a derived character within teleosts that may be termed convergent with that state in these primitive vertebrates.

Secondly, it is widely believed that individuals can have acellular bone in one section of the skeleton, and cellular in another, which would make acellular bone an unreliable character for phylogenetic analysis. However, the elopiform *Albula vulpes* (Linnaeus) is the only species in which both cellular and acellular bone in the skeleton has been reported from a series of 136 actinopterygians surveyed by Moss (1961b). The operculum and bony gill elements of *A. vulpes* are acellular, whereas the rest of the skeleton is cellular.

Thirdly, Moss explored the possibility that there are environmental explanations for bone type in particular fish groups. He expected marine species which live in a relatively calcium-rich environment to rely less on calcium stores in bone than freshwater fishes which live in a relatively calcium-poor environment (Moss, 1961b). This view was abandoned when it was concluded that higher teleosts, such as the true freshwater cichlids, were found to have acellular bone almost exclusively.

The paradentary is a bone found in the lower jaw of some Indo-Pacific atherinomorph or silverside fishes: the family Phallostethidae (Roberts, 1971; Parenti, 1984) and the monotypic genus *Dentatherina* Patten & Ivantsoff (1983). This bone was used by Parenti (1984) as a derived character to hypothesize the sister-group relationship of *Dentatherina* and the phallostethids.

The paradentary forms as a rod of cartilage in the lower jaw ligaments. It is ossified in many adult phallostethids, and may possess teeth (Parenti, 1984). The latter observation prompted a histological examination of the paradentary to determine whether it has a dermal toothplate or if in fact teeth attach directly to chondral bone.

The study provided the opportunity to review theories of the association between bone and teeth in teleost fishes. Fink (1981) described derived types of actinopterygian tooth attachment in terms of the failure of teeth to become completely mineralized, although bone type was not mentioned as a correlate of tooth type. In tetrapods, calcium regulation is dependent upon interaction between a parathyroid gland, which produces parathormone, and cellular bone, which acts as a calcium store. Teleosts do not have a parathyroid gland. Higher teleosts, with acellular bone, and lower teleosts, with cellular bone, have different mechanisms of calcium regulation. The character of acellular bone is correlated with incompletely mineralized teeth. The physiological explanation for this correlation might be found in the calcium regulation mechanism.

I suggest that correlation of derived characters, on physiological or other bases, becomes a focus of phylogeny reconstruction to work towards a comprehensive biological classification of bony fishes.

MATERIALS AND METHODS

Preserved specimens of the SE Asian phallostethid *Neostethus siamensis* Myers were re-fixed in 10% formalin and decalcified in 3% HCl in 70% EtOH. Both cross-sections and full frontal sections, approximately 16 μm thick, of adult males and females were stained with Ehrlich's haematoxylin and counterstained with picro-ponceau, according to the schedule of Humason (1979). The sections were photographed using an Ultrastar light photomicrography apparatus.

Additional comparative material of ontogenetic series of atherinomorphs was prepared according to the counterstaining method of Dingerkus & Uhler (1977). In this procedure, bone is stained red with alizarin red S, cartilage is stained blue with alcian blue, and flesh is cleared with the digestive enzyme trypsin. Additional clearing was carried out with KOH. Final preparations were stored in glycerin. Alcoholic specimens were used also for comparison. These specimens and the cleared and stained preparations were studied using a Zeiss SV8 stereomicroscope with drawing tube and photomicrography attachments.

The specific identification and museum catalogue numbers, including locality data, for all comparative material, and other information concerning these preparations, are available from the author.

For general comparative information on the histology of teleost bone, the reader is referred to two recent histological atlases: Yasutake & Wales (1983) on the salmonids, lower teleosts with cellular bone, and Groman (1982) on *Morone saxatilis* (Walbaum), the striped bass, a higher teleost with acellular bone.

STRUCTURE OF THE PARADENTARY

An ossified paradentary was first reported in phallostethids by Roberts (1971), who concluded that the bone, unknown then in any other group of fishes, is a neomorph. Patten & Ivantsoff (1983) reported calcified nodules in

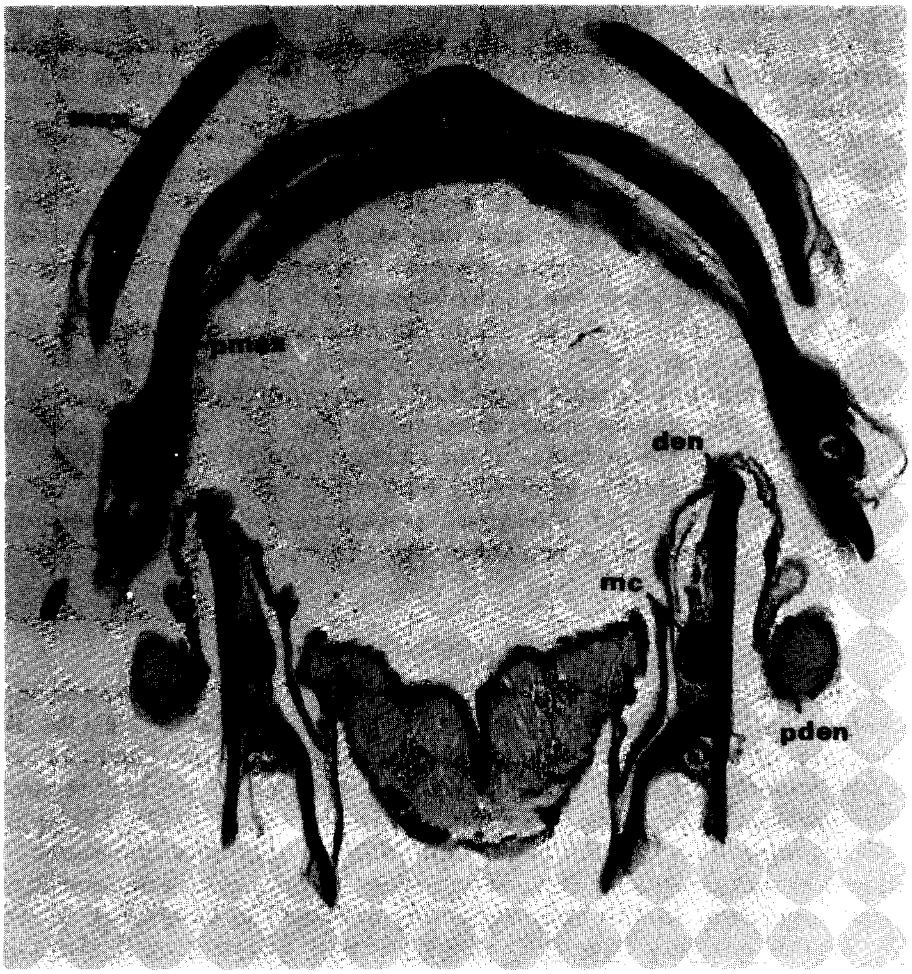
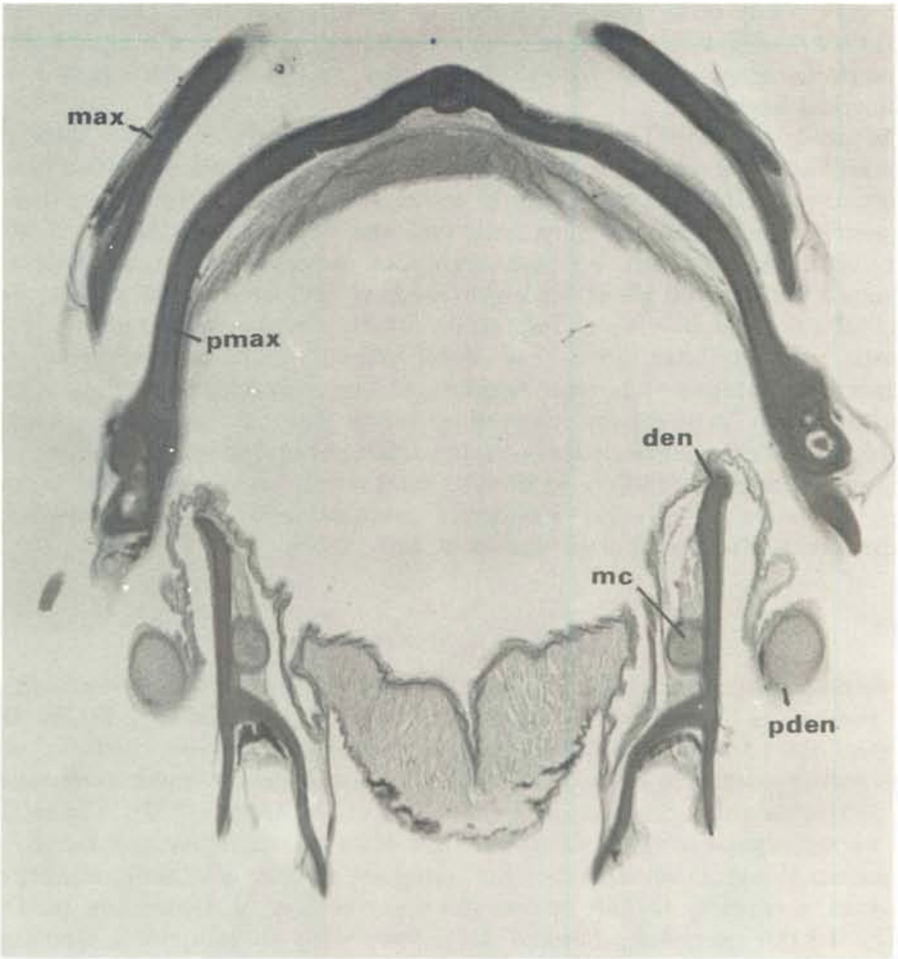


Figure 1. Histological section through anterior portion of the mouth of a male phallostethid, *Neostethus siamensis* Myers, stained with Ehrlich's haematoxylin, counterstained with picro-ponceau. den, dentary; max, maxilla; mc, Meckel's cartilage; pden, paradentary; pmax, premaxilla.

the lower jaw of *Dentatherina merceri,* a new genus and species of W Pacific marine atherinomorph, commenting that they were possibly like elements in the lower jaw of phallostethids. Parenti (1984) used the paradentary as a character to hypothesize the close relationship of *Dentatherina* and phallostethids, and reported that the paradentary in some phallostethid species may possess a row of small, unicuspid teeth. This observation, coupled with the fact that the paradentary forms with a cartilaginous precursor, prompted a histological examination of phallostethids to understand further the structure and formation of this lower jaw bone.

Phallostethids are small fishes, the largest adult body size recorded being just under 40 mm standard length (Roberts, 1971). The paradentary is a small, rod-shaped lower jaw bone that lies just lateral to the dentary (Parenti, 1984: fig. 1). In cross-section (Fig. 1) it appears similar to Meckels cartilage which lies just



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Phallostethus dunckeri Regan, a phallostethid from the Malay Peninsula, has a row of small, unicuspid teeth on the paradentary (Parenti, 1984). In other phallostethids with an ossified paradentary, such as *Neostethus siamensis* Myers, there are several teeth distributed randomly on the anterior portion of the bone. The teeth are incompletely mineralized; there is a ring of collagen at the base of each tooth. This structure is found in other teeth in the outer jaw, on the premaxilla and dentary, as well as on the pharyngeal arches. It may be referred to as Type 2 tooth attachment (as in Fink, 1981), a characteristic of other atherinomorph fishes.

Form and position of the paradentary corroborates the conclusion of Roberts (1971) that the bone is probably a neomorph in atherinomorph fishes.

No distinct dermal toothplate was observed, although this does not mean that one never exists during ontogeny. Dermal and chondral elements of teleosts may fuse during ontogeny. The resulting 'mixed bone' often contains hyaline cartilage and dense connective tissue, as well as bone (Groman, 1982).

Fusion between chondral and dermal bone may occur in ontogeny or phylogeny (Patterson, 1977). If fusion between the chondral paradentary and a dermal toothplate occurs in phallostethids, it is most likely a phylogenetic fusion; in no ontogenetic stage was a separate toothplate observed. Whether or not a toothplate exists does not affect the argument below that tooth type is correlated with bone type in teleost fishes. That no toothplate has been observed points to the possibility that higher teleosts may not necessarily follow rules of comparative anatomy that have been based on observations in higher vertebrates (see also Moss, 1961a; Lauder, 1983). Mechanisms for development of teeth on chondral bone are considered with regard to epidermal–mesodermal interactions in the section on bone, teeth and calcium regulation (pp. 43–46).

ACELLULAR BONE

That some teleosts have cellular bone, and others acellular, has been known for over a century, yet the statement by Moss (1961a: 102) that "Little attention has been paid to acellular bone" still pertains, at least with regard to use of this character in teleost phylogeny reconstruction.

Kölliker (1859a, b) presented the first systematic review of bone type in teleost fishes. He surveyed over 250 osteichthyans, and established the division of teleosts into those with cellular, and those with acellular bone. The one lungfish, chondrosteans (sturgeon and paddlefish), lepisosteid (gar), amiid (bowfin) and lower teleosts examined by Kölliker have cellular bone. Lower teleosts include osteoglossomorphs, elopomorphs, clupeomorphs, ostariophysans and the salmonids and argentinoids among Salmoniformes. Higher teleosts, which, for the purposes of this discussion, include Esocae (pikes, pickerels and mudminnows) and osmeroids (smelts and galaxioids) among Salmoniformes, and all neoteleosts, generally have acellular bone (for a comprehensive teleost classification see Lauder & Liem, 1983).

Several studies include reviews of literature on form and function of bone types (Enlow & Brown, 1956, 1957, 1958; Lopez, 1973; Schaeffer, 1977; Patterson, 1977; Meunier, 1984; and references therein). Moss (1961a, b, 1962a, b, 1963, 1965, 1968a, b, 1969, 1972) and co-workers (Moss & Posner, 1961; Moss & Freilich, 1963) addressed a range of phylogenetic, developmental, physiological and ecological questions concerning the form and function of acellular bone. The results of these studies need not be recapitulated here. I have drawn on these and other detailed experiments and discussions for an understanding of the unique aspects of acellular bone in higher teleosts.

The presence or absence of cells in scales or other exoskeletal features has not been surveyed as extensively as that in endoskeletal bone. Kölliker (1859b: 664) noted that in no fish whose endoskeleton lacks bone cells, do the cells exist in the hard structures of the skin; whereas those fish with cells in the endoskeleton may or may not have cells in such hard structures. Scales do act as important calcium stores, although the functional significance of this phenomenon has not been studied extensively (Weiss & Watabe, 1978, 1979).

Moss (1961b, 1965) continued the surveys of teleost species initiated by Kölliker. With refined histological techniques, Moss confirmed the division of teleosts established by Kölliker, and reported that one species, *Albula vulpes* (Linnaeus), has both cellular and acellular bone. Other elopiforms typically have cellular bone throughout the skeleton. Acellular bone in the operculum and gill elements may be interpreted as a secondarily derived character within *A. vulpes*. Other exceptions to the rule that lower teleosts have cellular bone and higher teleosts acellular are viewed tentatively as simply that (see the section on implications for euteleost phylogeny, pp. 46–48). For example, cellular bone of scombroids (Kölliker, 1859b) may represent a secondarily derived state within percomorphs.

What is considered to be more important for the classification of bone types is that there may be degrees of cellularity and acellularity within some species. For example, bone of salmonids is usually classified as cellular. However, Moss (1965) points out that number of bone cells varies within individual salmonids such that parts of the skeleton may actually be classified as acellular. This is of particular interest because the position of salmonids within a phylogeny of euteleosts is under debate (Rosen, 1973; Fink & Weitzman, 1982; Lauder & Liem, 1983; Fink, 1984). Salmonids may be characterized as intermediate between lower and higher teleosts, and their relative acellularity a stage in a transition series from cellular to acellular bone.

The position taken here is that acellular bone is derived relative to cellular bone, *not* because it occurs in higher teleosts, which would be a circular argument, but because it appears to be a derived state ontogenetically (although, for an alternative view, see Blanc, 1953). The organic matrix of teleost bone, composed of collagen and sulphated mucopolysaccharides, is calcified with a hydroxyapatite salt (Moss, 1961a; Moss & Posner, 1961). Both cellular and acellular bone have osteoblasts or osteocytes in early stages of development (Moss, 1961a; Schaeffer, 1977). In cellular bone these cells persist, as in the salmonids (Yasutake & Wales, 1983). In contrast, cells of mature acellular bone quickly become incorporated into the surrounding matrix. Cell death is rapid, and cell spaces become filled in, such that in histological section acellular bone has a relatively featureless matrix (e.g. Fig. 1; Moss & Posner, 1961; Schaeffer, 1977; Groman, 1982).

The obliteration of enclosed cells during acellular bone formation occurs as a result of intracellular accumulation of calcium salts, or intracellular mineralization (Moss, 1961a). Meunier (1984) performed quantitative microradiographic studies on 16 species of teleosts, those with cellular as well as those with acellular bone, and concluded that mean mineralization (measured as g hydroxyapatite/cm^3) is higher in acellular than cellular bone. Corresponding morphological characteristics of acellular bone are a significantly larger fraction of collagen and crystallites of smaller size than those of cellular bone (Moss & Posner, 1961; Moss, 1961a).

Physiological and phylogenetic implications of these characteristics of acellular bone, absence of cells and a larger fraction of collagen, are considered below for teleost fishes. Some aspects of these characteristics may be relevant to a study of the predominantly cartilaginous skeleton of chondrichthyans which can have acellular components (see above, and Peignoux-Deville & Janvier, 1984). However, even though an analogy between acellular skeletal components of chondrichthyans and higher teleosts may be drawn, these similarities are not viewed as homologous here.

BONE, TEETH AND CALCIUM REGULATION

An axiom of comparative vertebrate anatomy is that teeth are structurally similar to the bone with which they are associated. Moss (1968b) reminds us of the fundamental correlation between bone and teeth, postulating that dermal bone and teeth are 'homologous' structures. He does not mean homologous in the sense that teeth and bone are synapomorphous. By homologous, Moss implies a developmental and morphological correlation between teeth and bone, in particular dermal bone (in the sense of Patterson, 1977; see below).

It is not my purpose here to present a detailed comparison between the calcium-regulation mechanisms of teleosts and tetrapods, or to present a comprehensive review of hormonal control of mineral homoeostasis in teleosts, the focus of research by others (e.g. Pang, 1971; Chan, 1972; Pang *et al.*, 1978; Wendelaar Bonga, Van Der Meij & Pang, 1980). However, I point out major differences in the calcium-regulation mechanisms between lower and higher teleosts. Teleosts do not have a parathyroid, the primary calcium-regulatory gland in tetrapods, which produces parathormone. All gnathostomes have an ultimobranchial gland that produces the hormone calcitonin, a calcium inhibitor believed to have some calcium-regulatory function in teleosts (Copp, 1969; Wendelaar Bonga & Lammers, 1982). For example, when porcine calcitonin was administered to the eel *Anguilla anguilla* (Linnaeus) (in Copp, 1969) and the catfish *Ictalurus melas* (Rafinesque) (Louw, Sutton & Kenny, 1967), teleosts with cellular bone, plasma calcium levels were lowered. However, when calcitonin was administered to the killifish *Fundulus heteroclitus* (Linnaeus), a neoteleost with acellular bone, there was no change in plasma calcium level (Pang & Pickford, 1967). Wendelaar Bonga & Lammers (1982) corroborated this result for the cichlid *Oreochromis mossambicus*, concluding that (p. 60): "The absence of noticeable effects of calcitonin on the concentration of calcium and phosphate in bone and on plasma calcium may be related to the absence of osteocytes and the scarcity or absence of bone resorbing cells in acellular bone."

Osteocytes play a central role in mineral homoeostasis in teleosts (Moss, 1962a; Lopez, 1973). Initially, Moss (1961a, b) postulated that cellular bone was a character of freshwater fishes, whereas acellular bone was a character of marine fishes. A physiological explanation for this division was that freshwater fishes live in a relatively calcium-poor environment and need to rely more on an internal calcium store in bone than marine fishes which live in a relatively calcium-rich environment. However, Moss concluded that the type of bone had no correlation with environment but rather with phylogeny.

Moss (1962a) demonstrated that the absence of cells in mature bone of higher teleosts decreases the ability of the bone to function as a calcium reserve (see also Ellis, Roberts & Tytler, 1978). To test the effect of presence of cells on repair of fractures, hence movement of calcium or mineral homoeostasis, Moss (1962a) carried out a simple, yet pointed experiment. He broke the lower jaw and opercular elements of a goldfish, *Carassius auratus* (Linnaeus), a representative lower teleost with cellular bone, and a cichlid, *Tilapia macrocephala* (Bleeker) = *Sarotherodon melanotheron* Rüppel, a representative higher teleost with acellular bone.

Under normal diet and water conditions, both species responded to fracture by producing a large cartilaginous callus followed by calcified bone over the fracture site. This demonstrates that mineral homoeostasis is maintained in teleosts regardless of the presence or absence of cells in the bone (Moss, 1965). When the fish were reared in acalcemic water following fracture, the goldfish had the same response to the fracture as it had in normal water. However, the tilapia responded by producing a callus with poorly differentiated cartilage cells that remained uncalcified. That is, the goldfish could draw on the calcium store in its cellular bone, whereas the tilapia could not draw a comparable amount of calcium from its acellular bone.

It would be incorrect to say that teleosts with acellular bone are calcium deficient or that their bone is weak. However, teleosts with acellular bone cannot rely on calcium stores in bone as do teleosts with cellular bone, as well as all tetrapods.

Quite independent of these studies of bone, derived types of tooth attachment have been defined for actinopterygians in terms of failure of the tooth to become completely mineralized (Fink, 1981). A primitive tooth-attachment (Type 1) is a completely mineralized tooth fully ankylosed to the attachment bone. In derived attachment types teeth are successively unmineralized such that, for example in Type 2, there is a ring of collagen at the base of the tooth. Fink (1981) proposed Type 4 tooth attachment as a derived character of neoteleosts: there is a ring of collagen at the base, with a significant portion of this ring absent so that the tooth is hinged (for a discussion of tooth attachment types see Fink, 1981).

Incompletely mineralized teeth (Fink, 1981) and acellular bone (Moss, 1961a) share several characteristics, including a larger fraction of collagen. A physiological explanation for acellular bone supporting incompletely mineralized teeth is that the absence of cells precludes the bone from acting as a storage place for calcium (see above). Relative amount of calcium available in the bone for mineralization of teeth decreases as acellular bone develops ontogenetically. Thus, even though acellular bone is, on the average, more highly mineralized (Meunier, 1984), a major site of mineralization is the cell spaces, the filling of which limits internal calcium stores.

A dogma of comparative vertebrate anatomy is that teeth are associated only with dermal, and never chondral, bone. Schaeffer (1977) reviewed development of the dermal skeleton in vertebrates, concluding (p. 44): "... the calcified dermal skeleton in living fishes develops from a single modifiable morphogenetic system that is established by the interaction of the epithelium and the adjacent mesenchyme."

Analagous is Moss's postulate that dermal bone and teeth are homologous, that is, they are involved in a unified set of interactions between the ectoderm and the mesoderm. Patterson (1977: 79) defined dermal bone as that associated with "... ectodermal basement membrane by a surface coat of dentine and/or enameloid", pointing out that membrane bone, which has no developmental or phylogenetic connection with the ectoderm, should not be considered a synonym.

Patterson (1977) emphasized this distinction among bone types by reiterating that membrane and chondral bone are to be considered endoskeleton, whereas dermal bone is clearly to be considered exoskeleton. He proposed that (p. 86) "... all bones which are held to be homologous with tooth-, enameloid-, or sensory canal-bearing bones in primitive actinopterygians should be called dermal bones."

The structural unity between dermal bone and dental skeletal tissues was emphasized by Moss (1968b, 1969, 1972), who regarded these as constituents of the *integumental skeleton.* This system is not strictly homologous with the exoskeleton of Patterson (1977), who was not concerned directly with teeth, but perhaps is a more useful concept to describe the affinity between dermal bone and teeth (see Huysseune, 1983).

Moss (1968b) offered the *epidermal co-participation hypothesis* as a developmental explanation for this association. This theory of induction requires a relationship between, for example, the dermal skeleton and teeth simply because of the close association between these two tissue types and their developmental precursors. Tooth development necessarily requires this association because it is dependent upon induction in the mesoderm by overlying ectoderm.

That the paradentary lies lateral to the dentary presents the situation of close proximity of a chondral bone to the integument. Hence, we may think of the bone as an endoskeletal element in the position of the exoskeleton, to use the terminology of Patterson (1977). During ontogeny, conditions for tooth development are met. That is, presumptive bone cells lie under the ectoderm.

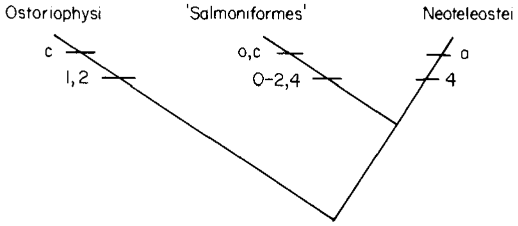

Figure 2. Cladogram of the groups of euteleost fishes, as proposed by Rosen (1973). 'Salmoniformes' indicate polyphyletic Salmoniformes. a, acellular bone; c, cellular bone. Numbers indicate predominant tooth attachment types, according to Fink (1981). See text for discussion.

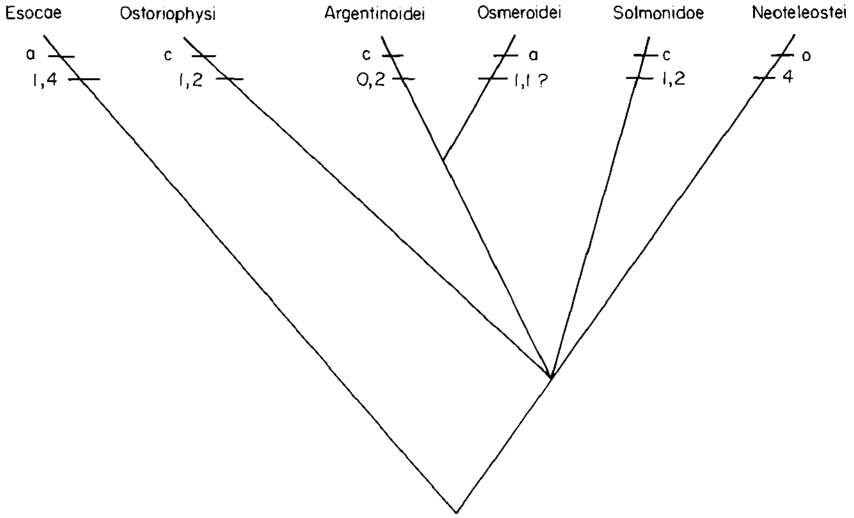

Figure 3. Cladogram of the groups of euteleost fishes, as proposed by Fink & Weitzman (1982). Ostariophysi and Neoteleostei have the same composition as those groups in Fig. 2. Esocae, Argentinoidei, Osmeroidei and Salmonidae constitute the Salmoniformes of Fig. 2. Abbreviations and numbers as in Fig. 2. See text for discussion.

That no dermal toothplate has been found on the paradentary does not affect, nor is it relevant to, this argument. Further, it is not proposed that this is reason to reject the current dogma that teeth are found generally on dermal bones. Rather, it is suggested that concepts such as that of the integumental skeleton are useful to define implications of the associations of developmental germinal layers, such as mesoderm plus ectoderm equals teeth, rather than more restrictive associations, such as presumptive dermal bone mesoderm plus ectoderm equals teeth. This is especially true for teleost fishes in which teeth are often widely distributed on skeletal elements that are associated with the integument during development. An example is the well-known clupeomorph *Denticeps clupeoides* Clausen, the skull roofing bones of which are covered with dermal denticles or odontodes (Greenwood *et al.*, 1966).

IMPLICATIONS FOR EUTELEOST PHYLOGENY

Teleost phylogeny has been the focus of much recent attention, particularly in the cladistic analyses of Fink & Weitzman (1982), Lauder & Liem (1983), Patterson & Rosen (1977) and Rosen (1973, 1982). The phylogeny of no other vertebrate group has been treated so extensively by cladistic methods. I argue that we must correlate many of the individual derived characters which have been used in these analyses to work towards a comprehensive biological classification of teleost fishes. Higher teleosts have a number of anatomical and cellular features that are unique among vertebrates, such as reduced cellular DNA content (Hinegardner & Rosen, 1972) and tooth attachment to bone via an unmineralized ring of collagen (Fink, 1981), or anatomical features with unique physiological properties, such as acellular bone (Kölliker, 1859a, b; Moss, 1961b; and discussion and references in the present paper). I propose that the last two of these, incompletely mineralized teeth and acellular bone, represent a character complex that is also correlated with the teleost calcium-regulation mechanism.

Fink (1981) characterized teleost evolution as paedomorphic because derived tooth attachment types of higher teleosts approximate earlier ontogenetic stages of the more primitive types. Acellular bone as an adult character represents a cessation of more typical bone cell development and may be considered degenerative. Reduced cellular DNA content may be correlated with these two characters, but it is unwise now to speculate on what a given amount of DNA means in teleost evolution (Hinegardner & Rosen, 1972). Reduced cellular DNA content, paedomorphosis or degeneration all may result in loss or reduction of structures.

Rosen (1973) presented the first comprehensive cladistic analysis of euteleost fishes (Fig. 2), concluding that the Salmoniformes are the sister-group of the Neoteleostei, and the plesiomorphic sister-group of both is the Ostariophysi. Monophyly of the last two groups has been corroborated (Fink & Fink, 1981; Fink & Weitzman, 1982). However, despite support for a monophyletic Salmoniformes (Rosen, 1974; Roberts, 1984), a definition of this large group of teleosts seems not to have withstood detailed analyses (Fink & Weitzman, 1982; Lauder & Liem, 1983). These authors did not mention acellular bone, but it too may be used as a character to reject salmoniform monophyly. The characters of cellular and acellular bone are indicated on the cladogram (Fig. 2) by a 'c' or an 'a', respectively. Ostariophysans generally have cellular bone, salmoniforms have cellular or acellular bone, and neoteleosts have acellular bone.

Patterson (1970) used the presence of acellular bone in two upper Cretaceous salmoniform genera to reject their placement in the Ostariophysi. He concluded further that the fossils were not argentinoids (salmoniforms with cellular bone) but may be more closely related to the osmeroids (salmoniforms with acellular bone). Patterson classified the genera as Salmoniformes *incertae sedis*, although he did not support strongly the monophyly of this order.

Fink & Weitzman (1982) recognized four groups in disbanding the salmoniforms: the Esocae (pikes, pickerels and mudminnows), the Argentinoidei (argentinoids) and Osmeroidei (smelts and galaxioids), and the Salmonidae (salmon, trout, whitefish and grayling). Their published cladogram (Fig. 3) does not agree with statements in their text concerning a possible sister-group relationship between the Salmonidae and the Neoteleostei (see Fink, 1984), but it is sufficient as an example in this discussion. Salmonids, like ostariophysans and argentinoids, generally have cellular bone, whereas Esocae, osmeroids and neoteleosts have acellular bone (Kölliker, 1859a, b; Berg, 1940; Moss, 1961b, 1965).

For groups recognized in both Rosen's (Fig. 2) and Fink & Weitzman's (Fig. 3) classifications, I have indicated type of tooth attachment by the numbers 0–4, following Fink (1981). The correlation between bone type and tooth attachment is imperfect, mainly because some species exhibit both primitive and derived tooth attachment modes, such as the chain pickerel, *Esox niger* Lesueur, in which outer teeth have Type 1 tooth attachment, whereas pharyngeal teeth have Type 4. That outer teeth have Type 1 attachment, i.e. are fully ankylosed to attachment bone, may be a characteristic of large predatory fish, and hence secondarily derived in higher teleosts. For example, the barracuda, *Sphyraena barracuda* (Walbaum), is a neoteleost with Type 1 tooth attachment in outer teeth and Type 4 in inner teeth. Fink (1981) treated the Esocae as the most primitive euteleostean clade, and hence not closely related to the Neoteleostei. He hypothesized that Type 4 tooth attachment in both *Esox* and neoteleosts was the result of homoplasy, and suggested that with more detailed study, development of the Type 4 tooth attachment in the two would be found to differ. However, given that, for example, *Esox* and *Sphyraena* have the same type of bone (acellular) as well as the same types of tooth attachment (Type 1 and Type 4), it is parsimonious to conclude that this similarity is derived.

It has been suggested by Lauder (1983) that prevailing cladistic hypotheses be used as the basis for interpreting functional morphological data within a phylogenetic framework. I point out that none of the character correlations presented here conflicts with Lauder's conclusions, which were based on treating Esocae as the most primitive of the euteleostean clades. Relationships of these clades are not known with certainty, and clarification of such is the subject of constant revision (Fink & Weitzman, 1982; Fink, 1984). I propose that acellular bone be treated as a derived character defining Esocae, Osmeroidei and Neoteleostei as a monophyletic group.

Lauder & Liem (1983) followed Fink & Weitzman (1982) in treating Type 4 tooth attachment as a derived character of the Neoteleostei. All neoteleosts do not have this type of tooth attachment, and they are not the only group in which this attachment is found. Nevertheless, I believe Fink & Weitzman (1982) were correct in postulating a major shift in tooth attachment type as a defining character of a large group of teleost fishes. I suggest that the Esocae and Osmeroidei be included in such a group, with an additional defining character being acellular bone.

CONCLUSIONS

Acellular bone, identifiable by absence of enclosed osteocytes in mature matrix, is a characteristic of higher teleosts, specifically the clades Esocae, Osmeroidei and Neoteleostei (Kölliker, 1859a, b; Moss, 1961b, 1965). Even though this character has been known since 1859, it is usually neglected in studies of teleost phylogeny. Knowledge of the developmental, physiological, ecological and phylogenetic aspects of acellular bone in teleost fishes, particularly that presented by Moss and co-workers cited herein, is extensive and should be incorporated into general theories of teleost phylogeny.

Derived types of tooth attachment in higher teleosts have been defined in terms of incomplete mineralization (Fink, 1981). Teleosts do not have a parathyroid, the major calcium-regulatory gland of tetrapods. Furthermore, the ultimobranchial gland, characteristic of gnathostomes, apparently does not have the same calcium-regulatory function in teleosts with acellular bone as it does in teleosts with cellular bone (Copp, 1969; Wendelaar Bonga & Lammers, 1982). Therefore, bone type is critical to the mechanism of calcium regulation in teleosts.

Moss (1968b) reminds us of the intimate relationship, in both structure and development, between bone and teeth. His concept of the integumental skeleton links exoskeletal and dental skeletal structures. I propose that the characters of acellular bone and incompletely mineralized teeth are a function of the calcium-regulation mechanism in teleost fishes, and hence should be correlated in comprehensive classifications. The implication of such a correlation is that these structures are part of a homologous system. By homologous, it is meant that these systems are developmentally and structurally unified.

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