

CHAPTER 14

An animal is a highly integrated machine and, because it is, it is convenient rather than analytical to regard it in pieces, as a collection of separate characters and adaptations.

Thomas H. Frazzetta (1975)

Integumentary, Sensory, and Visceral Systems

As was demonstrated in Chapter 13, the gestalt of an organism is determined by its musculoskeletal organization, which dictates how the animal can feed and move and, to a certain extent, determines its utilization of the physical environment. The musculoskeletal system, together with the integument, also protects and supports the soft anatomical components of the sensory, nervous, circulatory, respiratory, digestive, urogenital, and endocrine systems. The nervous system coordinates the activities of all other systems. Sensory structures are the receptors of environmental stimuli. Perceived sensory cues are transmitted to the central nervous system and thence to the peripheral and/or visceral nervous systems. Signals transmitted via the peripheral nervous system affect the musculoskeletal system directly, whereas those transmitted via the sympathetic (= efferent) and parasympathetic (= autonomic) portions of the visceral nervous system affect visceral organs of the circulatory, digestive, urogenital, exocrine, and endocrine systems.

The respiratory, digestive, excretory, exocrine, endocrine, and circulatory visceral systems are involved with the maintenance of the internal environment. Thus, metabolic requisites are introduced via the respiratory, integumentary, and digestive systems, and metabolic products are disposed of by the excretory, respiratory, and integumentary systems. Metabolic and endocrine products are transported throughout the body by the circulatory system to maintain a stable internal environment.

Only one visceral system is not involved in the maintenance of the internal environment; this is the reproductive system. Anatomically it is associated intimately with the excretory system; hence, the two usually are discussed as the urogenital system. The reproductive system acts in response to the internal environment and cues from the external environment, but its function obviously is to ensure the transmission of the parental genotype to a succeeding generation.

INTEGUMENT

Although the integument is the structural and functional interface between the organism and its environment, the morphological and functional complexity of amphibian skin is incompletely understood (see Lindemann and Voûte, 1976, and Whitear, 1977, for reviews). The skin of amphibians generally is described as being naked, that is, lacking the covering of scales, feathers, or hair characteristic of most other classes of vertebrates. Furthermore, amphibian skin is permeable to water and as such is important in respiration, osmoregulation, and to a limited degree, thermoregulation; these functional aspects are treated in Chapter 8. Also, the general appearance of amphibians is the result of integumentary structures; color and pattern are determined by the chromatophores, and texture is the result of integumentary modifications.

Structure

As in all vertebrates, the integument consists of an outer layer, the epidermis of ectodermal origin, and an underlying layer, the dermis. Most of the latter is of mesodermal origin, but the pigment cells are derived from the neural crest and thus are ectodermal; also, the glands imbedded in the dermis are derived from the ectoderm.

The outermost layer of the epidermis, the stratum corneum, consists of a single layer of flattened cells. The stratum corneum is keratinized in most adult amphibians, but it is not keratinized in obligate neotenic salamanders, such as *Necturus*. The keratinized stratum corneum is separated from the underlying stratum germinativum by irregular intercellular spaces that are interrupted by interconnecting filaments (desmosomes). The fibers of these keratinized cells form a double horizontal network reinforced by vertical bundles of filaments (tonofilaments) (Le Quang Trong and Bouligand, 1976) (Fig. 14-1). Underlying the stratum corneum is the stratum germinativum which normally is 4–8 cells thick; the innermost cells are columnar and the outer ones are progressively shorter. Lying within the stratum germinativum are specialized mitochondria-rich cells and flask cells of unknown function. The epidermis is separated from the dermis by a basement membrane of collagenous fibers.

The stratum corneum is sloughed (shed or molted) periodically. The duration of the intermolt period varies from 4–5 days in *Ambystoma* to 3–19 days in *Bufo* (Ling, 1972). In both salamanders and anurans, the stratum corneum splits middorsally beginning on the head; the

splitting of the dorsal skin progresses posteriorly. Most amphibians use their limbs to loosen and remove the slough either in patches or in one large piece; usually the slough is eaten. During the sloughing cycle, the intercellular subcorneal space between the stratum corneum and the underlying stratum germinativum is filled with mucus thought to be secreted by the mitochondria-rich cells. During actual sloughing, the desmosomal connections between the cells of the stratum corneum and the underlying replacement layer, derived from the stratum germinativum, are broken, and the desmosome fragments adhere to the sloughed stratum corneum. According to Budtz (1977), sloughing in *Bufo* is arrested by hypophysectomy; adrenocorticotrophic hormone (ACTH) and corticosteroids are the only hormones that elicit sloughing in hypophysectomized toads, but neither hormone has an effect on sloughing in normal toads. The formation of cocoons by aestivating amphibians is the result of multiple sloughs (see Chapter 8).

The dermis also consists of two layers. The outer stratum spongiosum is made up of areolar connective tissue with interlacing fibers and various types of cells, including the pigment-bearing chromatophores. The underlying stratum compactum is composed of compactly arranged collagenous fibers. Mucous and granular (= poison) glands of epidermal origin are imbedded in the stratum spongiosum, as are the scales in caecilians. Other structures in the dermis include capillaries, nerve fibers, and smooth muscles.

In salamanders and especially caecilians, there is a

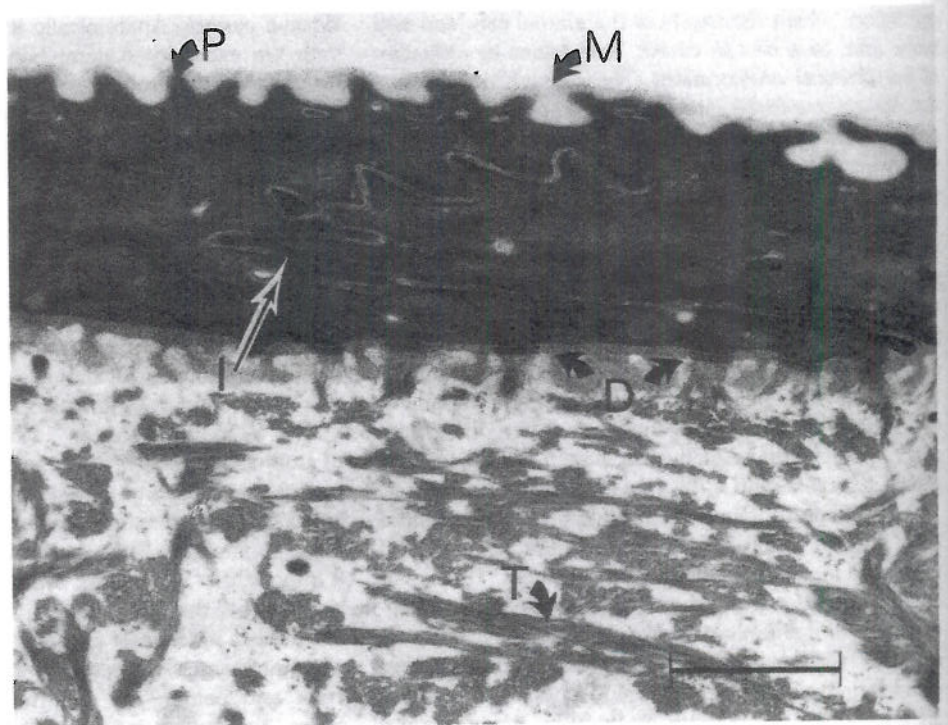


Figure 14-1. Electron micrograph of the surface layer of the epidermis of an anuran, *Phyllomedusa sauvagei*. D = desmosomes, I = intercellular junction, M = mucoid coat, P = protuberances on outer surface of epidermis, T = tonofilaments. Bar = 1 micrometer. Photo courtesy of R. Ruibal; reproduced with permission from *Copeia*.

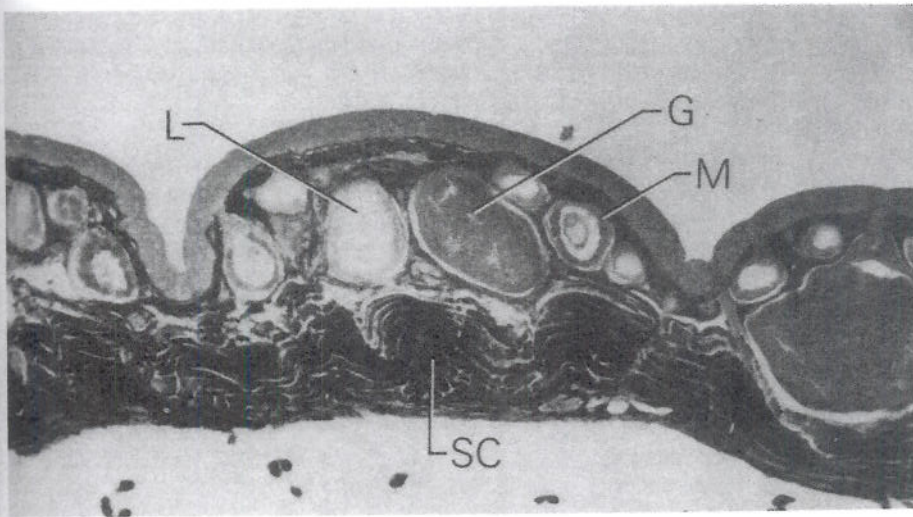


Figure 14-2. Photomicrograph of a vertical section of the ventral integument of an anuran, *Phyllomedusa sauvagei*. G = granular gland, L = lipid gland, M = mucous gland, SC = stratum compactum. Photo courtesy of R. Ruibal; reproduced with permission from Copeia.

practically imperceptible transition from the collagenous fibers of the stratum compactum of the dermis to the connective tissues covering the underlying bones and muscles. However, anurans are unique in having a loose skin attached to the body wall only at discrete places in one of the following ways: (1) by lymphatic septa which are thin, transparent sheets of connective tissue that divide the space between the skin and the muscles into separate compartments, the lymphatic sacs; (2) by fibers of transparent connective tissue commonly aggregated to hold a particular part of the skin close to the body wall; (3) by co-ossification of the skin with underlying dermal bones; (4) by direct attachment of the skin to muscles, as in the vocal sacs of some hylids (Tyler, 1971a); and (5) by cutaneous muscles that insert on the skin. T. C. Burton (1980) summarized previous work on cutaneous muscles and suggested that two cutaneous muscles, the m. rectus abdominis pars anteroflecta and the m. cutaneus dorsalis, may aid in the adoption of a defensive posture and in the secretion of fluids from integumentary glands in some microhylids.

Integumentary Glands. The epidermal glands imbedded in the dermis of amphibians have received considerable attention from morphologists. Important descriptive works on the glands in salamanders are those by Dawson (1920) on *Necturus*, Theis (1932) on *Salamandra*, and McManus (1937) on *Desmognathus*. Some earlier workers (e.g., Muhse, 1909) believed that only one kind of integumentary gland was present in amphibians, even in toads. It is now known that all amphibians have both mucous and granular (= poison or serous) glands.

Contrary views exist on the development of the glands. For example, Bovbjerg (1963) indicated that in *Rana*

pipiens the two types of glands and secretory cells develop independently without intermediate or transitional types. On the other hand, McManus (1937) found that the granular cells and glands of *Desmognathus fuscus* pass through a mucoid stage during histogenesis. During their development, some glands contain both mucus and granular material at the same time; intermediate stages could be traced from mucous to granular cells. In studies of *Ambystoma mexicanum* and *Nectophrynoides occidentalis*, Le Quang Trong (1966, 1967) noted that some glands have granular cells basally and mucous cells in or below the neck region.

Neuwirth et al. (1979) concluded that the granular glands are shared primitive characters among amphibians and their original function probably was other than poison synthesis, but the glands were a preadaptation for producing the diverse toxins that evolved separately in some groups of amphibians.

All of the glands are alveolar. Typical mucous glands are smaller than granular glands and enclosed completely in the stratum spongiosum (Fig. 14-2). In some salamanders and anurans, the mucous glands lack a distinct myoepithelium, but a distinct myoepithelium is present in caecilians (Fox, 1983). The bases of granular glands may project into the stratum compactum; the glands have one or two types of myoepithelial cells, and at least in some dendrobatids there is a layer of melanophores around the lateral and superficial surfaces of the glands (Neuwirth et al., 1979).

The numbers of mucous and granular glands vary throughout the body; generally mucous glands are more abundant in the dorsal skin than ventrally. Interspecific distribution of the glands, as shown in various ranids by Le Quang Trong (1971, 1975a, 1975b), may be related to differences in habitat. Also, mucous glands are more

numerous and widely distributed throughout the integument than are granular glands, which tend to be aggregated at specific sites in many species (e.g., head and neck of many anurans and some salamanders and dorsal surface of tail in other salamanders).

Mucopolysaccharides secreted spontaneously and continuously serve to keep the skin moist (see Chapter 8). Granular glands secrete only following sympathetic nervous or humoral stimulation. Various substances (e.g., peptides and alkaloids) in these secretions commonly are noxious and in some cases highly toxic; these secretions are important defense mechanisms (see Chapter 10). Three other kinds of integumentary glands are known in amphibians. Rather large, elongate glands are present in the skin in the dermal folds of caecilians; E. Taylor (1968) noted that these glands are associated with the dermal scales and suggested that secretions from these glands may form the scales. Blaylock et al. (1976) discovered lipid glands in the skin of *Phyllomedusa*, hylid frogs that secrete an impervious coating that protects them from desiccation (see Chapter 8). Lipid glands are slightly larger than granular glands, usually are in contact with the stratum corneum basally, and have a distinct myoepithelium. Breeding glands in the skin of the chest region of the microhylid *Gastrophryne carolinensis* were described by Conaway and Metter (1967); similar glands have been noted in other microhylids (see Chapter 3). The breeding glands are about the same size as the granular glands. The secretion is released by the fragmentation of the superficial part of the gland, and the sticky secretion adheres the male to the dorsum of the female. Histochemically, the secretion is similar to that of the mucous glands, but it lacks the sulfate groups characteristic of mucus (Holloway and Dapson, 1971).

Clusters of mucous or granular glands form obvious integumentary structures (macroglans) in many anurans and in some salamanders. Many of these structures develop only in males in response to testicular hormones; these structures are present only in the breeding season (see Chapter 3). The most widespread of these are nuptial excrescences, which are highly keratinized clusters of mucous glands, on the thumbs of many kinds of anurans and on the limbs of some salamanders. Clusters of granular glands may be present only in males (e.g., mental glands in plethodontid salamanders), and although these glands may become enlarged during the breeding season and therefore be affected by testicular hormones, they are not strictly seasonal in their presence. Other clusters of granular glands, such as the dorsal warts and parotoid glands of bufonids and some salamandrids, the lumbar glands of several genera of leptodactylids, the tibial glands of some myobatrachids and bufonids, and the dorsolateral and dorsal ridges of ranids, are permanent structures. The secretions of many of these macroglans are known to be important in defense against predators (see Chapter 10).

The secretions produced by the integumentary glands

include numerous complex biogenic amines and active polypeptides. Erspamer (1971) noted the presence of three groups of aromatic amines and five groups of polypeptides. The amines are:

1. Indolealkylamines, including 5-hydroxytryptamine (5-HT), which is present in most families and genera of amphibians, and *N*-methylated derivatives such as bufotenin and bufotenidine, which are found in pipids, leptodactylids, bufonids, hylids, ranids, and some salamanders.
2. Imidazolealkylamines known from *Leptodactylus labyrinthicus* and *L. pentadactylus*. Related histamines occur in several unrelated genera (e.g., *Leptodactylus*, *Taudactylus*, and *Litoria*).
3. Hydroxyphenylalkylamines, including leptodactylin known from various leptodactylids, and epinephrine and norepinephrine known from *Bufo*.

The active polypeptides include numerous toxins (see Chapter 10) and other, less toxic substances:

1. Eledosine-like polypeptides, such as physalaemin isolated from *Physalaemus*, phyllomedusin from *Phyllomedusa*, and uperolein from *Uperoleia*.
2. Bradykinin and bradykinin-like polypeptides, including bradykinin isolated from *Rana temporaria* and phyllokinin from *Phyllomedusa*.
3. Caerulein and caerulein-like polypeptides, including caerulein known from *Xenopus laevis* and various species of *Litoria* and *Leptodactylus*, and phyllocaerulein from *Phyllomedusa*.
4. Three types of alytesin and alytesin-like polypeptides: I from *Alytes obstetricans*, II from *Bombina bombina* and *B. variegata*, and III from *Rana pipiens*.
5. Miscellaneous polypeptides, including several other kinds, the chemical nature of which is not yet known.

The taxonomic distribution of many of these compounds generally corresponds to the classification based on other criteria, as noted for many South American anurans by Ceï and Erspamer (1966) and for Australo-Papuan anurans by Roseghini et al. (1976) and Erspamer (1984). Some groups of anurans (e.g., phyllomedusine hylids) contain large amounts of unique polypeptides, and each species has its own characteristic polypeptide spectrum (Ceï, 1963). The dendrobatids are well known for their strong toxins; *Phyllobates* produce mainly batrachotoxins, which are extraordinarily toxic steroidal alkaloids, whereas *Dendrobates* secretes a variety of less

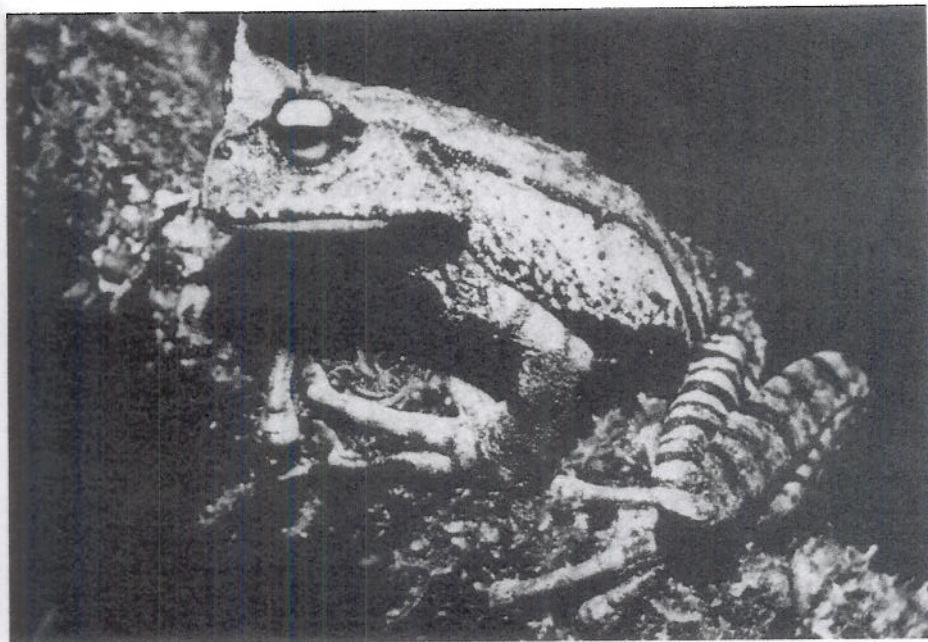


Figure 14-3. A hyloid frog, *Amphignathodon guentheri*, from Quebrada de Zapadores, Ecuador, showing supraciliary processes and calcars. Photo by W. E. Duellman.

toxic and chemically simpler piperidine alkyloids (Myers et al., 1978). Thus, the biochemical differences in the genera of dendrobatid frogs seem to have phylogenetic significance. Analyses of biogenic amines (Cei et al., 1972) and secretions of the parotoid glands (B. Low, 1972) of *Bufo* from throughout the world showed that trends in these biochemical traits corresponded to morphological groups of toads presumably representing different evolutionary lineages.

Too little is known about the factors affecting the biochemistry of granular glands to allow meaningful generalizations. For example, Myers et al. (1978) noted a decline in the toxicity of secretions produced by *Phyllobates terribilis* maintained in captivity. There even may be differences in the presence of a substance in secretions from glands on different parts of the body in the same species. Serotonin was identified in the secretions of parotoid glands of *Bufo alvarius*, but that compound was absent in secretions from the macrogland on the hindlimb (Cannon et al., 1978).

Texture and Integumentary Structures. Although the skin of many amphibians appears to be smooth, usually it has a texture owing to various dermal and/or epidermal modifications. Detailed macroscopic and histological studies of the amphibian integument by Rabl (1931) and H. Elias and Shapiro (1957) have shown that the epidermis varies in thickness and may have projections or indentations, and that there are elevations and thickenings of various kinds in the dermis, especially in anurans. H. Elias and Shapiro (1957) provided definitions and a terminology of the fine structures of anuran integ-

ument and noted that in many taxa, verrucae (warts) or conii (pointed projections) have the apex covered with keratin. These structures give a roughened, sandpaper-like structure to the skin.

Caecilians have dermal folds or annuli encircling or partly encircling the body. These annuli reflect body segmentation. Primary annuli overlie the vertebrae and myotomal septa, whereas secondary annuli (when present) lie between the septa (M. Wake, 1975). The costal grooves in salamanders also reflect body segmentation; the grooves overlie the myotomal septa and mark the position of the ribs.

Some integumentary structures, such as costal grooves in salamanders, granular ventral skin and lateral cutaneous channels in anurans, dermal flaps in some aquatic salamanders (e.g., *Cryptobranchus*) and anurans (e.g., *Telmatobius culeus*), and the hairlike projections on the flanks and hindlimbs of the aquatic ranid *Trichobatrachus robustus*, are associated with increased cutaneous vascularity. Increased surface area and vascularity function to increase water uptake in terrestrial amphibians, whereas increased surface area in aquatic amphibians provides for increased respiration (see Chapter 8).

Some kinds of integumentary structures seem to be associated with disruptive outlines and thereby aid in concealment (see Chapter 10). Such structures include small, irregular ridges, supraciliary processes, scalloped folds on the outer edges of limbs, and calcars (Fig. 14-3). The latter are elongated triangular flaps on the heels of some anurans. The presence of calcars in many kinds of arboreal frogs living in rainforests and their absence in other anurans invites the speculation that they

might serve as points for runoff of water, much the same as drip tips on leaves, or they may mimic drip tips.

Local thickenings of keratinized epidermis are present on the feet of various amphibians. The tips of the digits of some stream-dwelling salamanders of the families Hynobiidae, Ambystomatidae, and Plethodontidae have keratinized caps, and these are pointed and clawlike in the hynobiid *Onychodactylus japonicus*. Keratinized digit tips are present on the forefeet of *Siren*. Keratinized, pointed, clawlike tips also are present on the inner three toes of frogs of the genus *Xenopus*. The inner, and sometimes outer, metatarsal tubercles of several kinds of fossorial anurans (e.g., *Rhinophrynus*, *Scaphiopus*) are enlarged and covered with thick layers of keratin.

The webbing between the fingers and toes of anurans is entirely integumentary. Webbing commonly is absent on the hands; it is most extensive on the feet of many aquatic frogs (e.g., pipids, *Telmatobius*, *Pseudis*, and many *Rana*); in these frogs the extensive webbing obviously provides greater surface area for the feet in propelling the animal through the water. Several arboreal anurans (some species of *Agalychnis*, *Hyla*, and *Rhacophorus*) have fully webbed hands and feet. These frogs are capable of parachuting or gliding because of the great surface area present when the fingers and toes are spread (D. Davis, 1965); one species is able to attain a gliding angle of 55° (Table 14-1).

Also related to locomotion is the grasping ability of the toes of many kinds of arboreal frogs that have expanded adhesive toepads. Light and electron microscopical studies by Ernst (1973a, 1973b) and D. Green (1979) have shown that the epidermal cells in the toepads are structurally different from other epidermis on the body. The toepads of arboreal frogs of the families Hylidae, Hyperoliidae, and Rhacophoridae are nearly hemispherical structures on the ventral surfaces of the distal segments of the fingers and toes. The pad is bordered, except proximally, by a circumferal groove (transverse groove or circummarginal groove of some authors). The epidermal

cells of the toepad are columnar, usually hexagonal in shape, and clearly separated from one another at their apices (Fig. 14-4). The outermost surfaces of these cells are flat but covered with small, round hemidesmosome plaques. Epidermal cells elsewhere on the digits are squamous, except in an area of transition where the circumferal groove is absent; in this area the cells are cuboidal. In some frogs, cuboidal epidermis also is present on the subarticular tubercles. Interspersed among the columnar cells are mucous pores; these are numerous and bordered by unmodified cells in hyperoliids and rhacophorids, and less numerous and bordered by modified cells in hylids. The mucous glands imbedded in the dermis are large, convoluted, and surrounded by a thin myoepithelium of smooth muscle. The toepads are offset from the plane of the digit by an intercalary element between the distal and penultimate phalanges; this allows the entire surface of the toepad to be in contact with the substrate. Experiments by S. Emerson and Diehl (1980) and D. Green (1981) provided evidence that surface tension (capillarity) enhanced by mucous secretions is the principal means by which anurans adhere to smooth surfaces. Adhesion by toepads is supplemented by adhesion of the skin of the belly, also by surface tension. On rough surfaces, the structure of the epidermis allows interlocking of the toepad with the substrate.

Many species of the plethodontid salamander genus *Bolitoglossa* are arboreal and have thick interdigital webbing and shortened digits so that the hand and foot are padlike with a continuous smooth margin; the epidermis on the plantar and palmar surfaces is exceptionally smooth. Microscopical and experimental studies by Alberch (1981) and by D. Green and Alberch (1983) have shown that the principal mechanism of adhesion to smooth surfaces is suction created by the careful placement of the feet, the smooth perimeters of which adhere to the substrate while the middle part is lifted above the substrate. The suction requires moisture which is provided by the mucous glands in the palmar and plantar dermis.

Table 14-1. Results of Jumping Tests and Total Hindfoot Area of Gliding or Parachuting Frogs^a

Snout-vent length (mm)	Number of trials	Total area of hindfeet (mm ²)	Height of release (m)	Horizontal length of jump (m)	Angle of glide (degrees)
<i>Rhacophorus otitophus</i>					
86	2	53	5.4	3.0-4.0	30-38
72	1	39	5.4	3.7	35
<i>Rhacophorus pardalis</i>					
43	2	38	5.4	3.2	33
42	1	38	5.4	2.5	26
<i>Rhacophorus nigromaculatus</i>					
89	3	221	5.4	4.8-7.3	42-55
<i>Phrynohyas venulosa</i>					
—	—	—	42.7	27.4	34
<i>Agalychnis spurrelli</i>					
46-67	—	—	4.5	1.5-4.0	18-41
50 ^b	—	—	4.5	2.2 ^b	23 ^b

^aAdapted from N. Scott and A. Starrett (1974).

^bMedian values.

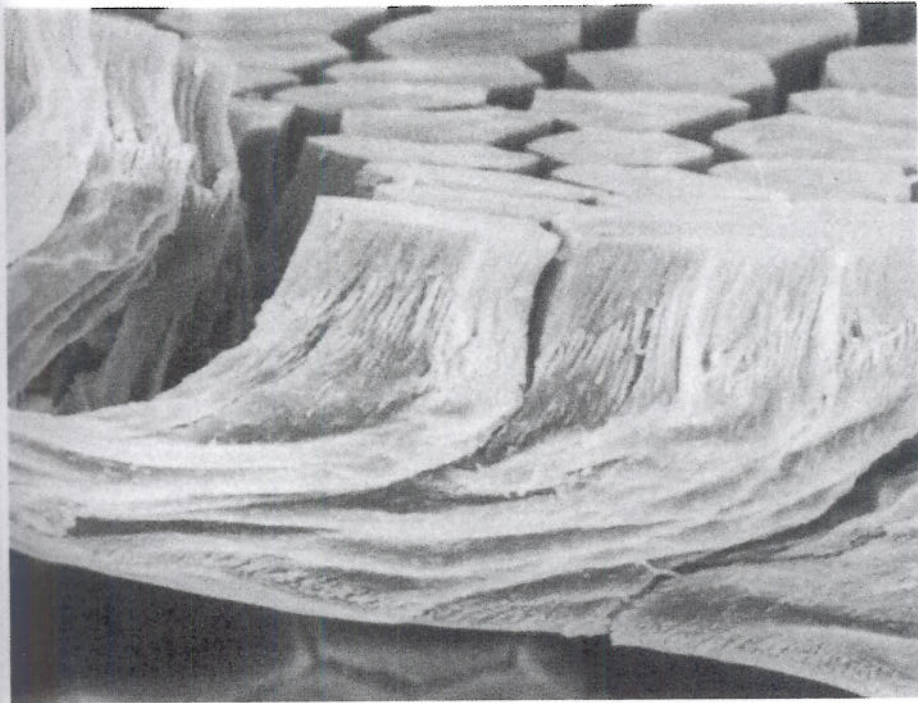


Figure 14-4. Scanning electron photomicrograph of the epidermal surface of a toepad of *Hyla versicolor*. Photo by D. B. Green.

The only other major modifications of the integument are associated with the brooding of eggs or tadpoles. The dorsal pouch in hylid marsupial frogs (*Gastrotheca*) is formed as an invagination of the integument (del Pino, 1980a). In comparison with the normal skin, the lining of the pouch is less keratinized and has numerous mucous glands. At the time of incubation the lining of the pouch becomes highly vascularized and forms partitions between the embryos. Eggs of some other hylids (*Cryptobatrachus*, *Hemiphractus*, *Stefania*) are carried openly on the back; mucous glands on the dorsum secrete a matrix that forms a pad to which the eggs adhere. The dorsal skin of female *Pipa* becomes thin prior to breeding; eggs adhere to the dorsum and sink into the skin, which subsequently thickens and encapsulates the embryo. The lateral pouches in which tadpoles of *Assa* develop are integumentary invaginations like those of *Gastrotheca*.

Dermal Ossicles and Co-ossification. The skin of several kinds of anurans contains bony structures. The dermis of the dorsal skin on the body of some hylid frogs (*Gastrotheca weinlandii*, *Phyllomedusa bicolor*, *P. vailanti*) contains small, vascularized bony plates (osteoderms) from which bony lamellar spines protrude into the epidermis. The pelobatid *Megophrys nasuta* and the leptodactylid *Hylactophryne augusti* have avascular osteoderms composed of calcified bundles of collagen. In all of these frogs the collagen fibers of the stratum compactum often are continuous with the ossified lower surface of the osteoderms (Ruibal and Shoemaker, 1984).

Brachycephalus and leptodactylids of the genera *Ceratotophrys* and *Lepidobatrachus* have large dermal plates in the dorsal skin (Fig. 14-5). These vacularized bony shields are separated from the epidermis by loose connective tissue, except that rugosities on the outer surface of the shield lie adjacent to the epidermis in *Brachycephalus*. In the latter, the plates of the shield are fused to the neural spines of the vertebrae.

The skin on the head is co-ossified with the dermal roofing elements of the skull in several groups of anurans, particularly in *Bufo* and several genera of hylids. In such cases, the stratum germinativum of the epidermis is compacted. The collagenous fibers in the dermis become ossified with the underlying bone, and the mucous glands and capillaries in the dermis are greatly reduced or absent, as the dermis becomes co-ossified with the underlying bones (Trueb, 1966). Co-ossification is associated with exostosis of the dermal bones (see Chapter 13).

Among amphibians, caecilians are unique in that many species have dermal scales; these are small, flat discs set in pockets in the transverse folds (= annuli). The scales arise in the pockets, and the basal part of each scale is attached to connective tissue in the deep part of the pocket. The scales have three principal layers. The basal layer is cellular. The middle portion is composed of bundles of parallel collagenous fibers arranged in two or three layers of different orientations. The superficial layer consists of mineralized squamulae separated from one another by concentric and radial furrows lacking mineralization. The most superficial of the fibrous layers is unmineralized and

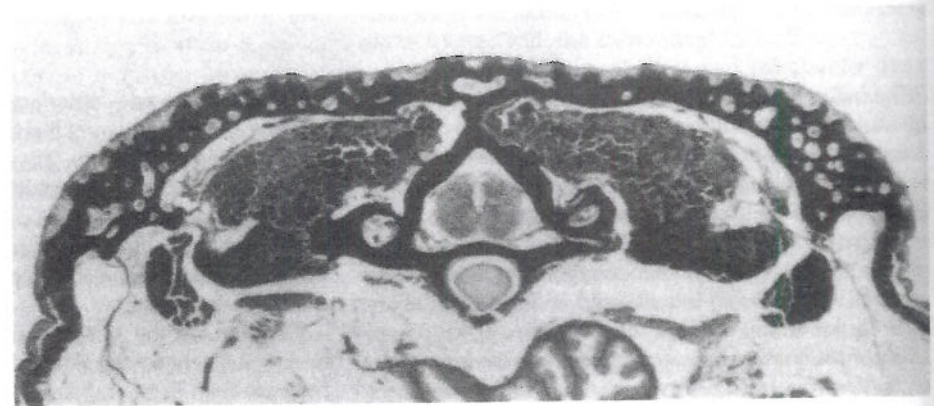
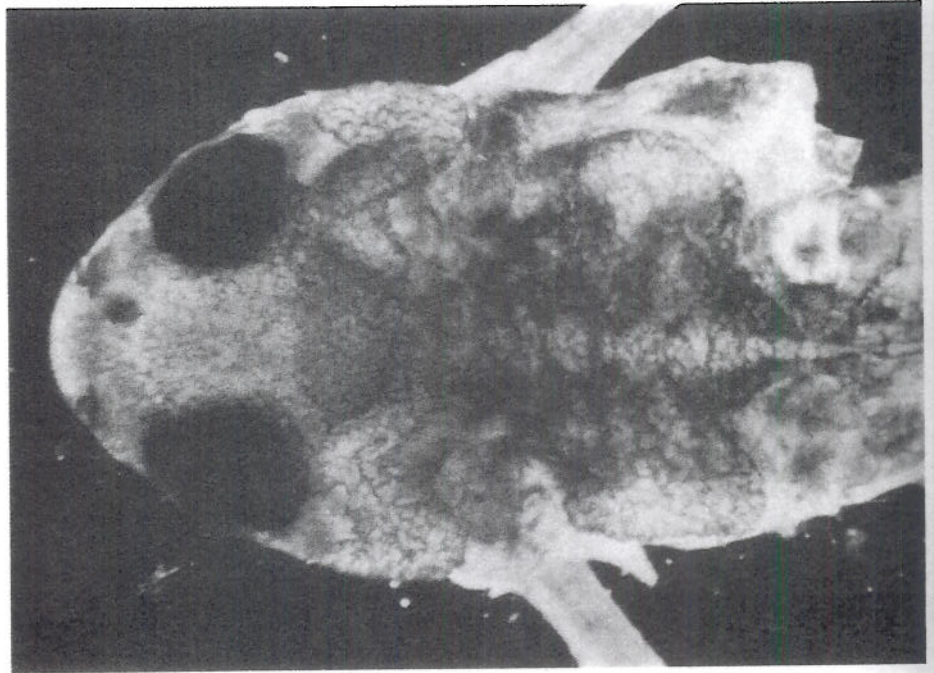


Figure 14-5. *Brachycephalus ephippium*. **A.** Dorsal view of cleared and stained adult (snout-vent length 16 mm) showing hyperossification of skull and presence of a broad dermal plate overlying vertebral column. **B.** Transverse cross section of midbody vertebra showing fusion of dermal spine with overlying dermal shield. Bones are black. Photos by L. Trueb.

is contiguous with the mineralized squamulae; mineral deposits are present on those fibers that extend into the squamulae (Zylberberg et al., 1980).

E. Taylor (1972) provided an atlas of caecilian scales in which he noted that the patterns of concentric and radial furrows were different among taxa. The question of independent origin of caecilian scales versus homology with osteichthyan scales is unresolved (see Zylberberg et al., 1980, for discussion). Although the scales may provide some protection, the large number of scales arranged in specific circular and overlapping repetitive patterns along the body and associated with the musculature may have a role in locomotion by providing requisite rigidity while permitting a wide range of body movements. Such a function might explain their usual absence in the aquatic typhlonectids, but scales also appear to be

absent in scolecomorphids and some caeciliids, both of which are fossorial.

Chromatophores and Pigmentation

The chromatophores and pigments in amphibians have been studied extensively, particularly by Bagnara and his associates—Bagnara and Ferris (1971), Bagnara and Hadley (1969, 1973), Bagnara et al. (1969, 1973, 1978, 1979), Hadley and Bagnara (1969), S. Frost and Bagnara (1979a, 1979b), and S. Frost and S. Robinson (1984). The synthesis that follows is derived primarily from these works.

Structure and Function. Amphibian chromatophores are located in either the epidermis or dermis. Melanophores are the predominant type of epidermal chro-

matophores, although erythrophores have been observed in the epidermis of some amphibians (e.g., *Notophthalmus viridescens*, Forbes et al., 1973). Epidermal melanophores are thin, elongate cells with long dendritic processes that extend between surrounding cells. For example, in tadpoles of *Bombina orientalis*, epidermal melanophores form an elaborate orthogonal network composed of melanophores each with four dendritic processes radiating symmetrically from a central cell body (Ellinger, 1980). Epidermal melanophores are characteristic of larval amphibians; upon metamorphosis these may be lost or their number reduced as the dermis thickens and dermal chromatophores develop. Epidermal melanophores, like those in the dermis, produce pigments known as eumelanins, which are deposited within organelles known as melanosomes. An epidermal melanophore unit consists of a melanophore and surrounding epidermal cells (Malpighian cells) that act as receptors of melanin donated by the melanophores through cytokine activity.

In the dermis of most amphibians that have been studied there usually are three types of chromatophores that are arranged in what has been termed a dermal chromatophore unit (Fig. 14-6). In this unit, xanthophores (or erythrophores) are the most superficial cells; they lie just below the basement membrane separating the epidermis and dermis. Xanthophores impart yellow, orange, or red colors primarily because of the presence of pteridine pigments. In some cases the red, yellow, or orange colors observed in amphibian skin are caused by the presence of carotenoid pigments that are concentrated in carotenoid vesicles in cells best described as erythrophores. Some anurans, such as *Bombina orientalis*, have chromatophores that contain both pteridines and caro-

tenoids in discrete pigment organelles (Fig. 14-7A). Such cells are best termed xantho-erythrophores.

Iridophores underlie xanthophores; these cells (also called guanophores) are white or silvery in appearance and have the capacity to reflect light of specific wavelengths through the overlying xanthophores. Together, the xanthophores and iridophores interact to produce bright colors. Iridophores reflect light by virtue of the arrangement of their pigment-containing organelles (i.e., reflecting platelets). These organelles commonly are arranged in parallel stacks which thus function as a multi-layer interference reflector (i.e., a "mirror," S. Frost and S. Robinson, 1984). The principal pigments in these cells are purines (e.g., guanine, hypoxanthine, adenine).

Often, the iridophore layer in amphibian skin is only one cell layer thick. An exception occurs in the arid-adapted species of African rhacophorid frogs, *Chiromantis*, which have chromatophore units containing 3–5 layers of iridophores; presumably this results in increased reflectance, which might be correlated with reducing evaporative water loss (Drewes et al., 1977). In a blue morph of *Dendrobates pumilio*, xanthophores are lacking and iridophores are stacked in layers above the underlying melanophores (Fig. 14-7B; S. Frost, unpubl. data). In this case the iridophores reflect blue light, which, in the absence of overlying red or yellow pigments, imparts a structural blue color to the skin.

Melanophores are the basal-most chromatophores; dendritic processes extend upward to terminate on the upper surfaces of iridophores, between these cells and the overlying xanthophores. The principal pigment of dermal melanophores is eumelanin. In phyllomedusine and some pelodyadine hylids (Bagnara and Ferris, 1975;

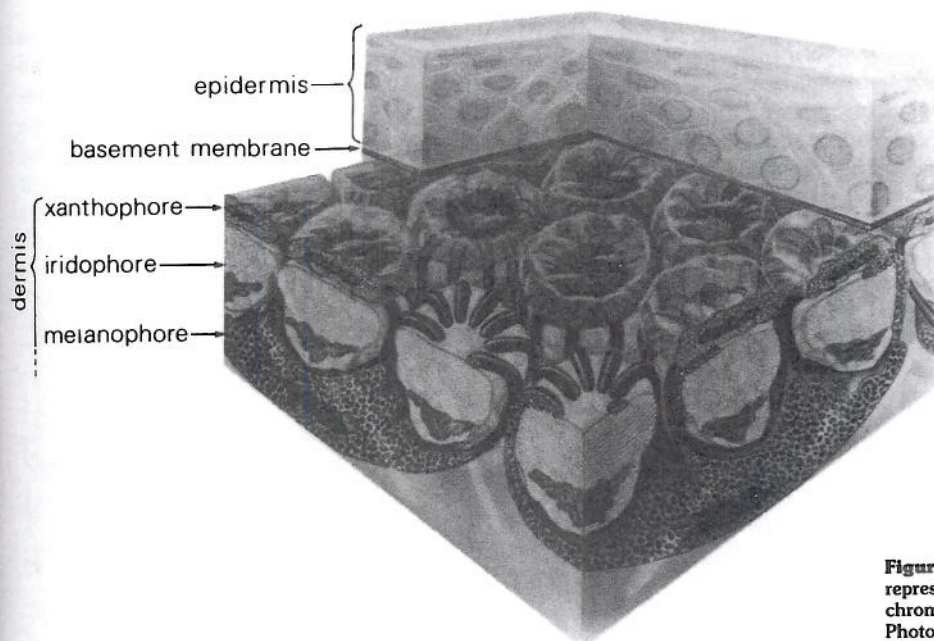


Figure 14-6. Diagrammatic representation of dermal chromatophore unit in a dark phase. Photo courtesy J. T. Bagnara.

Tyler and M. Davies, 1978b), melanosomes are extremely large and contain, in addition to a eumelanin core, a red pigment, pterorhodin, which is a pteridine dimer. This pigment and unusual type of melanosome are unknown in other vertebrates (Misuraca et al., 1977).

Characteristically in adult amphibians, the dermal chromatophore unit responds to physiological changes by affecting a change in color, usually manifested as a darkening or lightening of the skin. It is only when dermal chromatophore units contain two or more chromatophore types, with melanophores residing deepest within the unit, that color changes are affected.

In amphibians, blue skin is primarily a structural phenomenon resulting from both the reflecting and scattering (i.e., Tyndall scattering) properties of iridophores and the absence of xanthophores or bright-colored xanthophore pigments. When yellow pigments are present in the xanthophore layer, the pigments act as filters such that blue light reflected from the underlying iridophores and passing through the yellow layer will appear green. This, in fact, is the basis for the observed green skin color of many amphibians, although shades and intensity of green colors may vary tremendously.

According to S. Frost and S. Robinson (1984), there are three major factors contributing to green color in frog skin: (1) the kind of yellow pigment (i.e., carotenoids or pteridines) in xanthophores, (2) the quantity of yellow pigment in xanthophores, and (3) the arrangement of pigment organelles in the iridophores below the xanthophores. The first two factors presumably account for differences in the intensity of green color, whereas the last probably affects the quality (shade or tone) of green. In their studies of *Bombina orientalis*, these investigators noted that the structural arrangement of reflecting platelets in the green dorsal skin is such that these organelles function as refractosomes (i.e., multiple-layer interference reflectors). Furthermore, the red color on the venter of *Bombina* is explained easily because ventral skin lacks iridophores, only a few melanophores are present, and the xanthophores contain only carotenoid vesicles. The pigment in these vesicles is densely concentrated carot-

enoid which appears red-orange. Thus, differences in location and composition of chromatophores result in the different colors and patterns observed in amphibians (see reflectance model of Nielsen and Dyck, 1978).

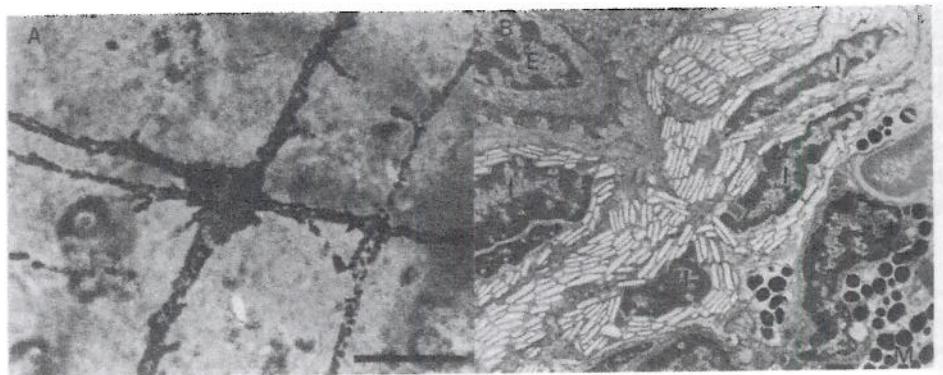
In addition to the presumably genetically based color dimorphism observed in many species of amphibians, there are occasional examples in nature of melanism and albinism (Dyrkacz, 1981). The genetic basis for the color variants has been studied in species of *Rana* and in the axolotl, *Ambystoma mexicanum* (C. Richards and Nace, 1983; S. Frost et al., 1984, and references therein). Peculiar blue variants are known in *R. catesbeiana* and *R. clamitans*. The problem in these frogs seems to reside in the xanthophores, which have abnormal pterinosomes and lack bright-colored pteridine pigments (Bagnara et al., 1978).

Bagnara, Matsumoto et al. (1979) proposed that the different chromatophore types are derived from a common stem cell containing a primordial organelle. Cues present in the tissue milieu dictate the fate of this organelle. Thus, differentiation of the stem cell into a specific chromatophore may be controlled genetically, hormonally, or environmentally. Developmentally, pigmentary changes may occur at various stages in the life cycle. C. Richards (1982) determined that chromatophore pigments change ontogenetically with the development of sex hormones in the African treefrog *Hyperolius viridiflavus*. During and subsequent to metamorphosis, the majority of pterinosomes in xanthophores are replaced by carotenoid vesicles in the dorsal skin of *Bombina orientalis* (S. Frost and S. Robinson, 1984) and *Notophthalmus viridescens* (Forbes et al., 1973). Thus, the chromatophores and pigment compositions of the amphibian integument represent a highly dynamic system.

Color Change. Two kinds of color change can be affected by the chromatophore units of amphibians. Rapid color changes involving intracellular mobilization of pigment-containing organelles are referred to as physiological color changes; this change results from hormonal stimulation. These changes may require only seconds to

Figure 14-7. Photomicrographs of amphibian chromatophores.

A. Epidermal melanophore pattern in dorsal skin of a tadpole of *Bombina orientalis*. The melanophores form a highly organized, reticulate network within the epidermis. Bar = 25 μ m.
B. Stacked iridophores in the dermis of *Dendrobates pumilio*. The skin of this specimen was a deep blue-black. Two layers of reflecting pigment cells (iridophores) are located immediately below the epidermis. Melanophores lie beneath the iridophore layer. Abbreviations: E = epidermal cell, I = iridophore, M = melanophore process. Bar = 1 μ m. Photos by S. K. Frost.



accomplish and commonly are of short duration (minutes to hours). Color changes that are evoked slowly and that involve the accumulation or reduction of the amount of pigment are referred to as morphological color changes. This is a slow process because it involves the synthesis or destruction of relatively large amounts of pigment as a result of either the persistence or continuous lack of stimulation of the chromatophores. Such changes are of long-term duration (days to months).

Epidermal melanophores undergo morphological changes. Some anurans subjected to continuous stimulation deposit so much melanin in adjacent epidermal cells that practically the entire epidermis becomes melanized. Within the dermal melanophore unit, increases in melanin may be accompanied by a decrease in guanine in iridophores. Thus, the morphological effects elicited by iridophores and melanophores are supplemental to one another.

Morphological color changes usually are preceded by physiological color changes, but morphological color change is not a necessary consequence of physiological color change. Morphological color change involving an increase in the amount of pigment contained in a chromatophore seems to be related to the dispersion of the pigment-containing organelles in the cell, just as a decrease in pigment content is accompanied by an aggregation of the organelles in the middle of the cell. The occurrence of morphological color change in the absence of physiological change was noted by J. D. Taylor (1969) for *Hyla cinerea*; when the frogs were treated with MSH (melanocyte-stimulating hormone; intermedin) there was diminution of purines in the iridophores, but the organelles remained aggregated.

Physiological color changes in the dermal chromatophore unit involve the dispersion and aggregation of melanosomes and reflecting platelets. In a pale color phase, melanosomes are aggregated in a perinuclear position, and the melanophores contribute little to the color of the animal. Concomitantly, the iridophores are not obscured by overlying melanin and the reflecting platelets are fully dispersed (in those iridophores that show physiological color change). Xanthophores, which apparently do not undergo cellular rearrangement during physiological color change, play only a passive role by serving as a yellow filter. Thus, the net effect of these iridophore and melanophore responses is that the animal becomes paler in color. In a dark color phase, there is an aggregation of iridophore pigments thereby reducing the reflecting surface area and a dispersion of melanosomes into the melanophore projections that cover the iridophores.

Nielsen's (1978a) ultrastructural studies of color change in *Hyla arborea* also demonstrated that during the change from a pale to dark color phase: (1) dermal melanophore projections partly surround the xanthophores as well as the iridophores, (2) iridophores change from a cup shape to a cylindrical or conical shape, with a simultaneous reorientation of platelets from being parallel to the upper

surface to being irregular, and (3) xanthophores change from a lens shape to a plate shape. During intermediate darker stages, xanthophores migrate down between iridophores and may even go beneath iridophores; the pterinosomes gather in the periphery of the cell, and carotenoid vesicles aggregate around the nucleus.

Color change in adult amphibians seems to be mostly, if not exclusively, controlled by circulating levels of MSH which disperses melanosomes in melanophores and causes aggregation of reflecting platelets within iridophores and dispersion of pigmentary organelles in some xanthophores. The secretion of MSH is under inhibitory regulation, possibly by the hypothalamus. Electrophysiological studies by Oshima and Gorbman (1969) indicated the presence of two types of active electrical units in the pars intermedia of anurans; one of these neurons is inhibited by light, and the other is indifferent to illumination. Thus, it was proposed that these two nervous elements are in balance and regulated by the influence of light on one of them. The light-inhibitable neuron is considered to stimulate the release of MSH under conditions of low illumination.

Amphibian larvae become pale when subjected to prolonged darkness; the melanosomes aggregate in the middle of the melanophores. Experiments by Bagnara (1960) revealed that this change was negated by pinealectomy, but administration of pineal hormones induced the aggregation of melanosomes. The hormone melatonin, secreted by the pineal gland, is a melanosome-aggregating agent. The release of melatonin is regulated by light receptors in the pineal body.

Controversy exists whether color change in adult amphibians is caused solely by levels of MSH or involves other hormones. In vitro, the contraction of iridophores by MSH can be counteracted by the administration of norepinephrine or acetylcholine (Bagnara et al., 1969). Nielsen and Dyck (1978) observed differential dispersion of organelles in the three types of chromatophores in *Hyla cinerea* and concluded that MSH could not be the only agent influencing color changes; they did not rule out the possibility of nervous control of the chromatophores, as did Bagnara and Hadley (1973). Moreover, chromatophores may be sensitive to light, as shown in *Pachymedusa dactylos* by Iga and Bagnara (1975).

Observations on numerous kinds of amphibians indicate that color change can be affected by changes in illumination and also by temperature. Moreover, in at least some species color change is affected by background color; the animals become darker on dark backgrounds. However, this is not a generality. For example, Nielsen (1979) experimented with two color phases of *Rana esculenta*; the green color phase changed in response to a pale background, whereas the brown phase did not. Experiments with *Hyla arborea* and *H. cinerea* by Nielsen (1978b) revealed that both species became pale when treated with epinephrine; also, stress (frogs pressed into a plastic box) affected the hue and purity of

the dorsal color of *H. arborea* and the paleness of *H. cinerea*.

The dilemma expressed by Bagnara and Hadley (1973:74) remains unresolved: "... chromatophores may be affected by either hormonal or neurohormonal agents as well as by direct environmental influences. . . . Certain hormones may be inhibited from being released under conditions of illumination, whereas others may be released under conditions of darkness, or vice versa. In either situation the chromatic responses may appear similar, although their regulatory basis may be quite different."

Integumentary Sensory Receptors

As the interface between the organism and its environment, the integument receives stimuli that must be trans-

mitted to the brain so that the organism may respond appropriately.

Lateral-line System. The lateral-line system is a collection of epidermal sense organs distributed over the head and along the body in aquatic amphibians. Lateral-line organs are present in aquatic larvae, aquatic adult salamanders (e.g., *Cryptobranchus*, *Necturus*, *Amphiuma*, and *Siren*), adult pipid frogs, and adult salamandrids that are aquatic after a terrestrial stage (e.g., *Notophthalmus*) or remain aquatic (e.g., *Neureergus*).

The biology of lateral-line receptors in salamanders and anurans was reviewed by Russell (1976) and in caecilians by Hetherington and M. Wake (1979). Also, new information was provided on salamanders (*Neureergus*) by Gorgees et al. (1977). Earlier work on the lateral-line system involved only the identification of mechanoreceptors, the neuromasts. The identification of ampullary organs that are electroreceptors has been more recent—Hetherington and M. Wake (1979), Fritzsche (1981), Münz et al. (1982), Himstedt et al. (1982), and Fritzsche and Wahnschaffe (1983).

The structure and arrangement of the lateral-line organs (neuromasts) are similar in different amphibians. The organs are distributed singly (*Siren*) or in small groups along the lateral or dorsolateral surface of the body and especially on the head, where distinct patterns are evident on dorsal, lateral, and ventral surfaces (Fig. 14-8).

Each neuromast consists of a pear-shaped group of cells imbedded in the epidermis and resting on the basement membrane. Each organ is constructed of three types of cells: (1) mantle cells forming the periphery, (2) supporting cells internal to the mantle cells and extending from the basement membrane to the surface, and (3) sensory cells or hair cells in the apical half of the organ. The organs protrude beyond the surface of the skin in larvae but usually are recessed in a small pit in adults. The apical surface supports a thin, ribbonlike projection, the cupula. On the apical end of each sensory cell is a single, long kinocilium and a group of much shorter stereocilia, which decrease in length with increasing distance from the kinocilium (Fig. 14-9). Neuromasts are innervated by one efferent and two afferent nerve fibers. Most neuromasts on the head are innervated by the anterior lateral-line nerve (lateralis anterior of C.N. VII), and all remaining neuromasts by the posterior lateral-line nerve (lateralis posterior of C.N. X). Fibers from these two nerves enter the area of the acousticolateralis in the dorsolateral wall of the medulla.

In *Notophthalmus viridescens* the neuromasts are normal during the aquatic larval and adult stages, but during the subadult terrestrial stage the cells in the neuromasts dedifferentiate until they are alike cytogenetically (Dawson, 1936). In the adult aquatic stage, the sensory cells are replicated by division of both sensory and supporting cells. It is not known if there are changes in the nerve fibers innervating the neuromasts of *Notophthalmus*, but in the terrestrial stage of *Triturus cristatus*, which under-

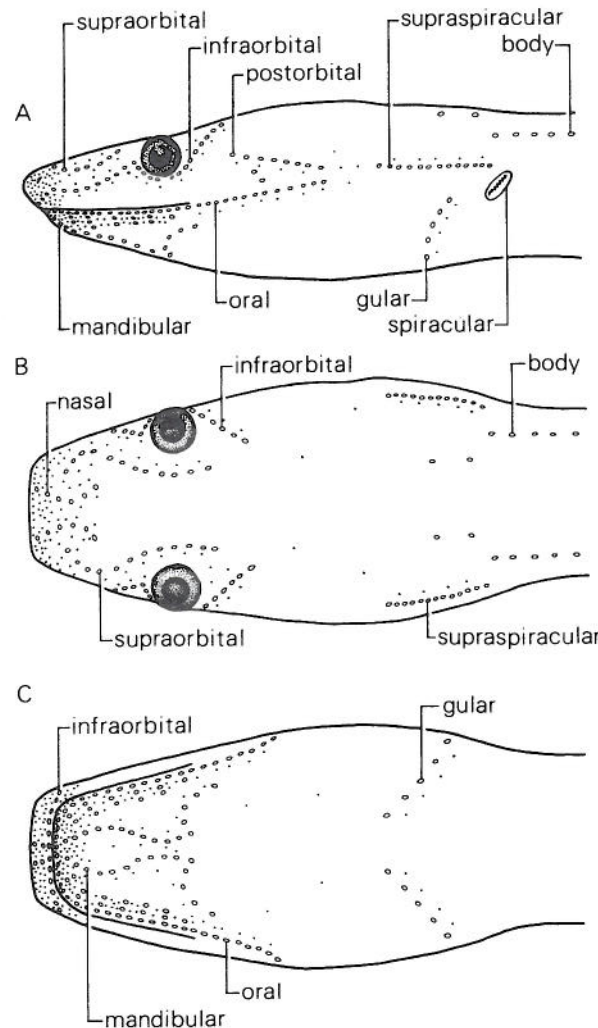


Figure 14-8. Distribution of lateral-line organs in *Ichthyophis*. A. Lateral. B. Dorsal. C. Ventral. Open structures are neuromasts; solid structures are ampullary organs. Redrawn from Hetherington and M. Wake (1979).