

# Distinct Mechanisms Underlie Pattern Formation in the Skin and Skin Appendages

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Patterns form with the break of homogeneity and lead to the emergence of new structure or arrangement. There are different physiological and pathological mechanisms that lead to the formation of patterns. Here, we first introduce the basics of pattern formation and their possible biological basis. We then discuss different categories of skin patterns and their potential underlying molecular mechanisms. Some patterns, such as the lines of Blaschko and Naevus, are based on cell lineage and genetic mosaicism. Other patterns, such as regionally specific skin appendages, can be set by distinct combinatorial molecular codes, which in turn may be set by morphogenetic gradients. There are also some patterns, such as the arrangement of hair follicles (hair whorls) and fingerprints, which involve genetics as well as stochastic epigenetic events based on physiochemical principles. Many appendage primordia are laid out in developmental waves. In the adult, some patterns, such as those involving cycling hair follicles, may appear as traveling waves in mice. Since skin appendages can renew themselves in regeneration, their size and shape can still change in the adult via regulation by hormones and the environment. Some lesion patterns are based on pathological changes involving the above processes and can be used as diagnostic criteria in medicine. Understanding the different mechanisms that lead to patterns in the skin will help us appreciate their full significance in morphogenesis and medical research. Much remains to be learned about complex pattern formation, if we are to bridge the gap between molecular biology and organism phenotypes. **Birth Defects Research (Part C) 78:280–291, 2006.** © 2006 Wiley-Liss, Inc.

## INTRODUCTION TO PATTERN FORMATION

What is a pattern? Patterning can be considered as the loss of homogeneity, when small, random perturbations to a system are amplified through a number of local processes and iterations to form recognizable structure or order (Meinhardt, 1982; Ermentrout and Edelstein-Keshet, 1993; Murray, 2003; Chuong et al., 2006a). For example, one of the simplest forms of patterning is the

asymmetric conversion of part of a homogenous field (Fig. 1A; gray) to a different state (Fig. 1B; black). The new pattern can be generated as dots, stripes, patches, segments, branches, etc. (Fig. 1C–E), and can be arranged randomly or periodically.

What are the mechanisms of biological pattern formation? In some cases, they may be based on the distribution of cell lineage so that cells strictly follow their fates genetically (Fig. 1F). In other cases, it

may be based on combinatorial molecular coding which can be interpreted at the enhancer/transcription factor level (Small and Levine, 1991) or at the cell adhesion level (Steinberg, 1996) (Fig. 1G and H). These molecular changes usually appear before the real morphological changes and are referred to as prepatterns (Nagorcka and Mooney, 1992; Forgacs and Newman, 2005). They can explain many downstream phenomena that follow the generated prepattern, but do not explain the upstream issue—we do not know how these molecular codes are set up. For example, morphogenetic gradient models have been proposed in which cells can interpret their position within a morphogen gradient, as Wolpert (1969) has proposed in the French flag model (Fig. 1C). Cells can enter a new state in a concentration dependent manner (Fig. 1I and J) (Ashe and Briscoe, 2006). This can explain many examples of how molecular codes are set, but still does not resolve the issue as to the origin of the pattern—we still do not know what sets up the molecular gradient, for example, how the exact morphogen and its point of secretion are determined. This is where self-organization comes into play: stochastic events combined with physi-

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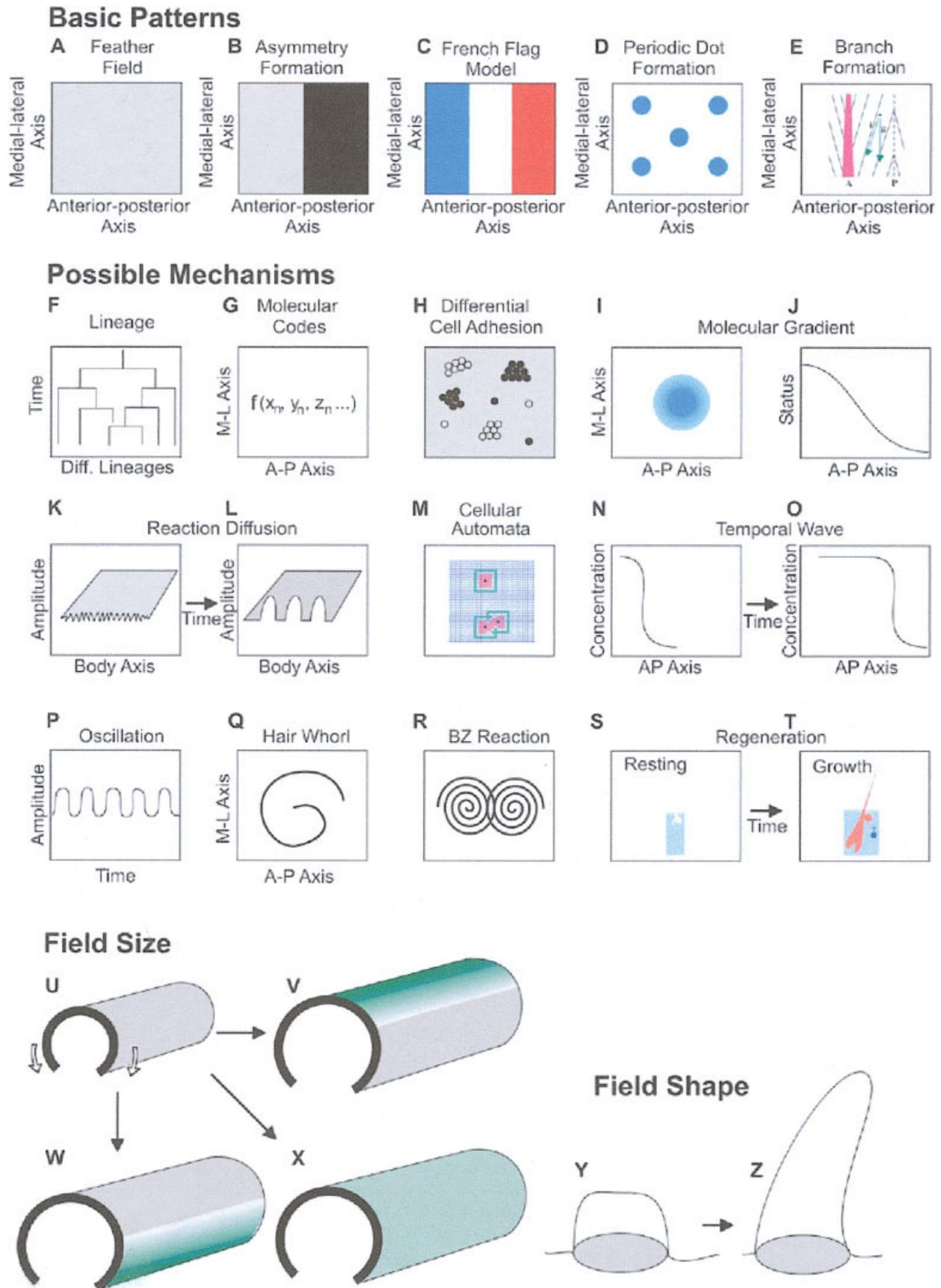
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**Figure 1.** Basics of pattern formation. **A-E:** Schematic drawings showing basic patterns. **F-T:** Possible mechanisms that can lead to pattern changes. **U-Z:** Additional factors that can influence biological pattern formatting due to growth. E: From Yue et al. (2005). M: From Jiang et al. (2004). U-X: Represent the trunk with the midline on the top. There are three possible ways new cells can be added, which are indicated by the green color. Y,Z: Indicate the changes of field shape which can represent the growth of limb bud, tail bud, or feather bud. See text for further explanation.

cochemical principles can increase the order and/or structure of a system, perhaps resulting in emergent events, without being guided by an external source (Newman and Comper, 1990; Camazine et al., 2003; Newman et al., 2006). In other words, patterns at the global level may solely arise as a result of interactions between lower level components.

Reaction diffusion models following the method first outlined by Turing (1952) have been applied to explain many biological periodic patterning processes (Fig. 1K and L) (Gierer and Meinhardt, 1972). In this model, the morphogenetic field starts with a homogenous distribution of cells, activators, and inhibitors, and random fluctuations initiate the periodic patterning process. The activators and inhibitors undergo a series of interactions, which can include self- and cross-activation and inhibition; both can diffuse, with the inhibitor diffusing further than the activator. With time, patterns in the form of dots or stripes in activator and inhibitor concentration gradually emerge, with the pattern depending on the ratio of activators to inhibitors and the size and shape of the pattern field. This mechanism has been proposed to explain the formation of hair follicles (Nagorcka and Mooney, 1992) and feather patterning (Jung et al., 1998).

Cellular automata models have also been proposed to explain many biological phenomena (Fig. 1M) (Wolfram, 1992; Deutsch et al., 2004). In a general cellular automata model, the field is divided into a number of discrete "cells," which evolve through a number of time steps, according to a set of rules based on the states of neighboring "cells." Each "cell" of the model corresponds to an area of the pattern field and information on this area is stored as the "state" of the cell. Along this line, a digital hormone model has been developed to explain the formation of dermal condensations by feather mesenchymal cells (Shen et al., 2004). The role that stochastic interactions may play in hair cycling have been explored using a cellular automation model (Halloy et al., 2002).

Oscillation is another important property that may be used in patterning. The oscillation can occur at the level of a single cell, or at the level of an organ (e.g., hair and feather follicle) (Fig. 1P, S, and T). A clock and wavefront mechanism involving cellular oscillation has been used to explain the formation of somites (Fig. 1N–P) (Pourquié, 2003). Oscillation of hair follicles in hair cycling becomes very visible in darkly pigmented normal mice, such as C57BL6/J (e.g. chnemus et al., 2005) and in mutant mice, such as nude mice and *Msx2* null mice (Militzer, 2001; Ma et al., 2003; Suzuki et al., 2003; Mecklenburg et al., 2005). A model based on the Belousov-Zhabotinski reaction was recently suggested to explain this phenomena of wave formation, although no underlying molecular basis was identified (Fig. 1R; Fig. 5) (Suzuki et al., 2003).

Of course, the physicochemical events are still genetically based since the biological patterns are species-specific. A way to conceive this is that DNA gives rise to RNA and proteins that build cells with unique physicochemical properties. At this level, groups of cells interact with outcomes based on these properties and the surrounding environment—not just on the molecule itself. Therefore, the pattern formation process is best appreciated as a combination of genetic and epigenetic events, and the results are both deterministic and stochastic, as seen in the fingerprints of homozygotic twins: similar but nonidentical (reviewed in Jiang et al., 2004).

Organs can also grow and change their shape, size, and organization during development (Fig. 1S and T). Another level of variation is that the morphogenetic field (in this case, the surface of the animal body) changes in size and shape during development (Fig. 1U–X). The way in which these changes take place can influence patterns which are at a formative stage. For example, during the expansion of the skin, new dermal and/or epidermal cells may be added to specific regions such as those receiving cells from the dermatome or the advancing ventral

body folds. In other cases, new cells may be inserted randomly all over the developing skin (Fig. 1U–X). These growth modes can have different consequences for patterns forming on the skin. Similarly, the shape of the morphogenetic field may change, even sometimes reducing in size, and this also may lead to variation in patterning; for example, the formation of stripes rather than spots (Fig. 1Y and Z) (Murray, 2003).

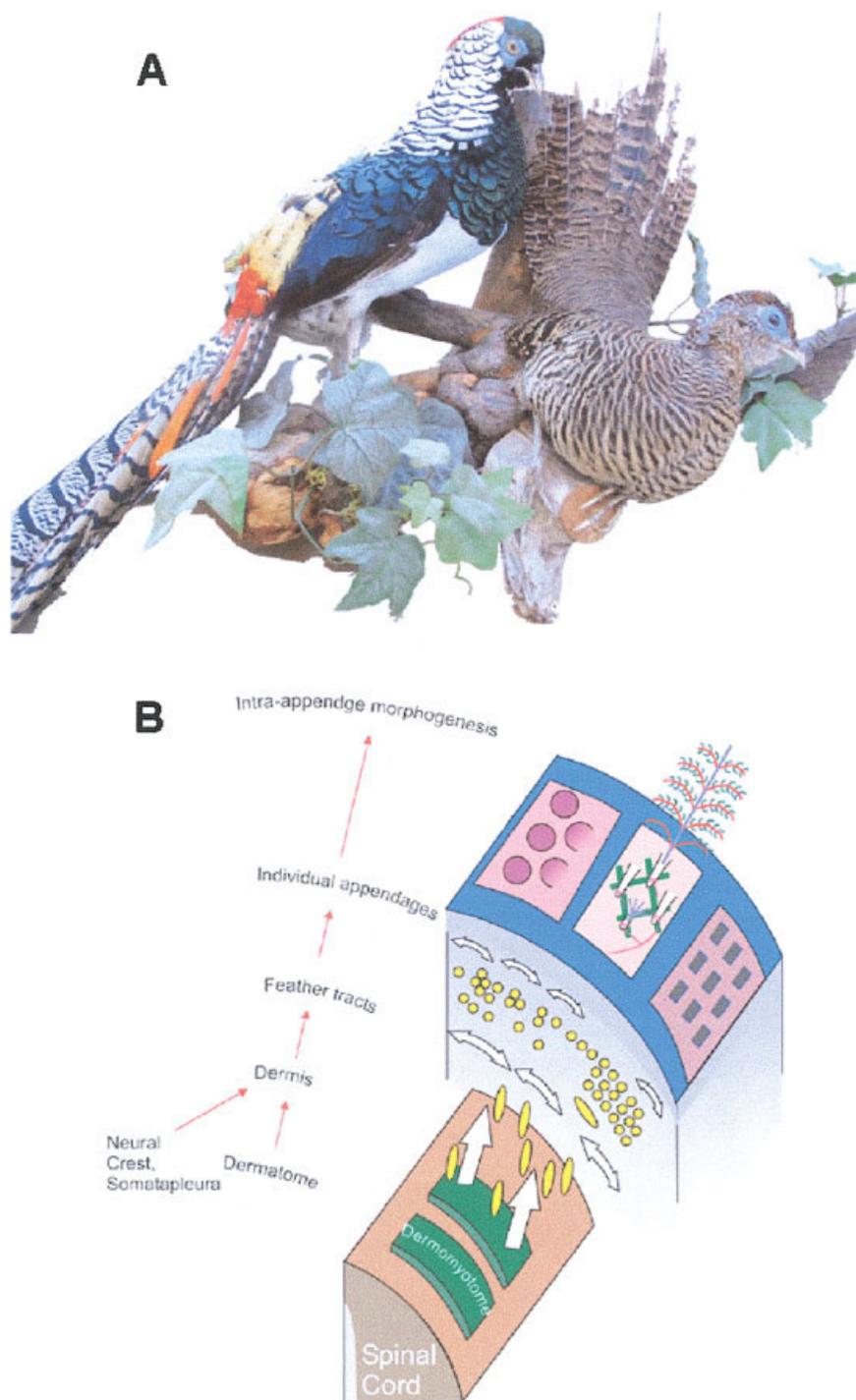
Our aim is to try to analyze these pattern formation processes and identify the biological bases underlying them, but much remains to be learned. We have described above only some examples, and they certainly do not exhaust all models that have been proposed for patterning. We can also contemplate that complex pattern formation is generated through a combination of the above processes, which perhaps results in patterns that are more robust to genetic and environmental perturbations. We will point out these patterns and the processes that may be involved in the following sections.

## DEVELOPMENT-BASED AND UNALTERABLE PATTERNS

Although the appearance of integuments of mammals, birds, and reptiles can be very different (Fig. 2A), the development of their skin and skin appendages share similar hierarchical morphogenetic processes (Fig. 2B). In some cases, different types of skin appendages appear, while in others, the patterns of similar types of appendages are arranged differently (Ball, 1999). We think that this is controlled through genetic and epigenetic controls that operate at different levels (Jiang et al., 2004). Here we take a closer look at these regulatory processes.

### Regional Specificity

Regional specificity implies that different skin regions such as the scalp, beard, eyebrows, face, lips, palms, nails, mammary glands, sweat glands, etc., have different characteristics. Epidermal precursors (or stem cells)



**Figure 2.** Patterns on avian skin and skin appendages and hierarchical morphogenesis. **A:** Male and female pheasants show regionally specific skin appendages and sexual dimorphism. Also note the thick pigment stripes and dots in the tail feather. Prum and Williamson (2002) proposed a reaction diffusion model for feather pigment patterning. **B:** Different developmental stages of skin appendage morphogenesis. Note the different types of skin appendages, including the schematic radially and bilaterally symmetric feathers. Modified from Wu et al. (2004).

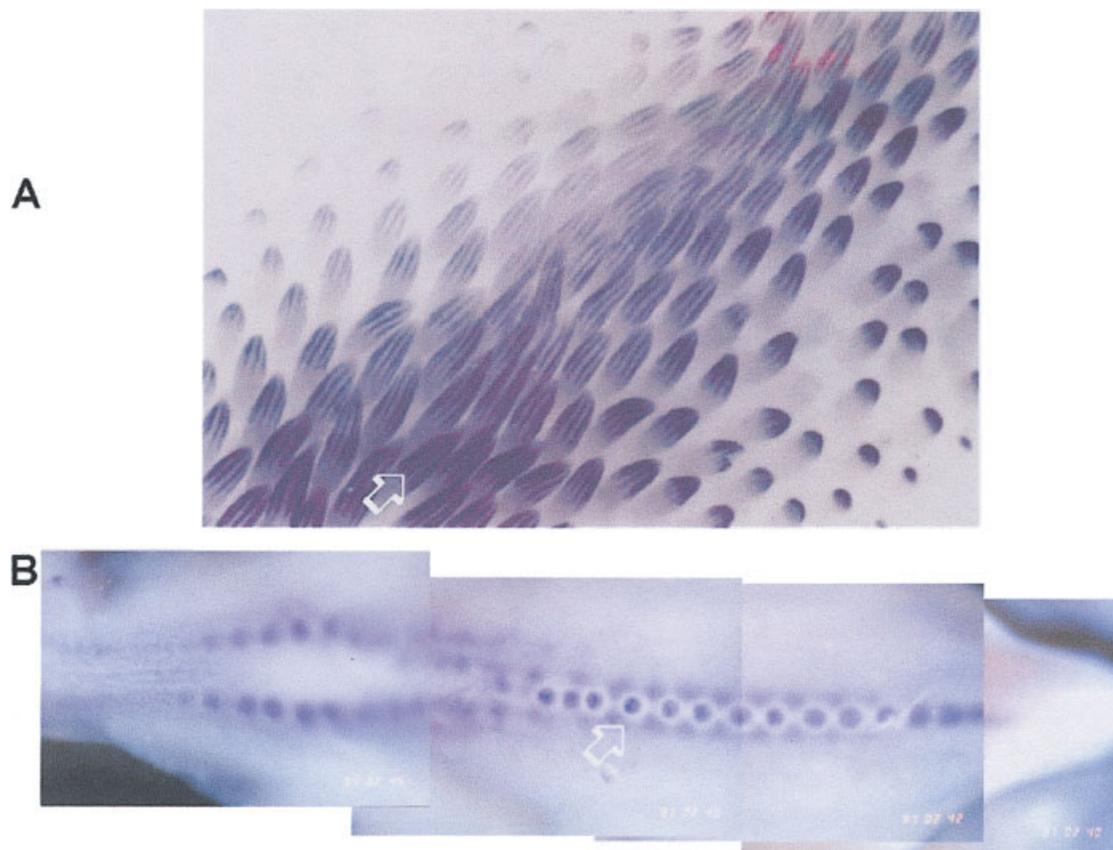
are initially multipotent and competent to form all these different structures. During development, special domains of the dermis begin sending

specific messages to the epidermis. Through a series of epithelial–mesenchymal interactions, these different skin domains with special structures

and functions gradually emerge. The integument diversifies to endow different functions to different parts of the human skin. An example of regional specificity can be seen by comparing the different types of feathers present on the breast, wing, tail, etc., of birds. This is most evident in pheasants, as shown in Fig. 2A.

How dermal specificity and epidermal competence are set up is mostly unknown. A model based on a skin Hox code was proposed, suggesting that different combinations of Hox gene expression may be the basis of skin regional specificity, setting up the subsequent differences in diffusible morphogens and adhesion molecules (Chuong, 1993). Different Hox expression patterns are shown in different regions of chicken skin (Kanzler et al., 1997; Duboule, 1998; Reid and Gaunt, 2002). Indeed there are spatiotemporally defined, specific HOX expression patterns in human skin (Stelnicki et al., 1998), and the Hox expression patterns of dermal cells derived from different topological skin regions in humans are different (Chang et al., 2002).

Most interestingly, the characteristics of these different skin regions can be trans-determined. For example, the engrailed pathway was shown to be involved in defining the mesenchymal characteristics of the ventral versus the dorsal paw (Loomis et al., 1996). *Tbx4* and *Tbx5* are shown to be involved in defining the identity of the chicken leg versus wing, and hence scale or feather forming dermis (Rodriguez-Esteban et al., 1999). Epidermal cells can trans-differentiate and convert hairs into glands or scales into feathers under the influence of retinoic acid, or by ectopic expression of specific molecules such as  $\beta$ -catenin (Dhouailly et al., 1980; Robinson et al., 1990; Widelitz et al., 2000). A recently engineered K14-noggin transgenic mouse shows that sweat glands are transformed to hairs (Plikus et al., 2004), while noggin overexpression under the neuron-specific enolase promoter can convert outer root sheath keratinocytes into sebocytes (Guha et al., 2004). An adult cornea can



**Figure 3.** Temporal wave. **A:** In situ hybridization of Shh in embryonic chicken skin. Midline is indicated by an arrow. Feather bud formation starts from the midline, and then lateral buds appear sequentially. From the lateral edge to the midline are regions of no feather primordia, feather placodes, short buds, long buds, and feather filaments with branch formation. **B:** In situ hybridization of  $\beta$ -catenin. The feather field first homogeneously expresses  $\beta$ -catenin at a moderate level in the morphogenetic zone. Then the periodically arranged buds emerge gradually expressing high levels of  $\beta$ -catenin, while the lateral inhibitory zone does not express beta catenin. From Widelitz et al. (2000).

also be diverted to form pilosebaceous units when they are confronted with embryonic hair forming dermis (Pearson et al., 2004). These observations imply that the specific combinatorial molecular codes may specify phenotypes of skin and skin appendages (Chuong, 1993; Fliniaux et al., 2004; Prin and Dhouailly, 2004). Altering these codes may lead to a resetting of the phenotypes. The upstream question concerning how the molecular codes are set remains unanswered.

### Developmental Wave

During skin development, hair or feather primordia are laid out in a temporal order as they gradually acquire competence (reviewed in Dhouailly et al., 2004). Their arrangement and orientation are reflected as a propagating wave of skin ap-

pendage formation. This process is clearly shown in the chicken skin in Figure 3A. In the spinal tract of the chicken, the formation starts at the midline and spreads bilaterally. At the lateral edge, feather primordia are at the induction stage. Toward the midline, they progress to form short feather buds, long feather buds, and feather filaments with branching morphogenesis, etc. A morphogenetic wave sweeping from the midline to the lateral has been inferred (Sengel, 1990). However, although the lateral row appears after the more medial row, the formation of the lateral row does not really have to depend on the medial row (Jiang et al., 1999). While this sequential appearance may be perceived as a "gradient," it is actually a temporal wave since the lateral feather buds will eventually also go through feather bud and filament stages. For a tract,

there has to be a primary row before this sequential appearance takes place. The gradual emergence of buds along the primary row in the midline can be readily visualized by in situ hybridization staining for  $\beta$ -catenin (Fig. 3B). The molecular basis of these process remains unknown.

In humans, this is manifested as hair whorl patterns in the occipital region (Fig. 1Q) (reviewed in Plikus and Chuong, 2004). In human fetuses, lanugo hairs form whorl patterns both on the scalp and trunk skin (Gworys and Domagala, 2003). On the thoracic wall there are lanugo whorls that begin bilaterally over the nipples. The whorls collide and merge along the midlines (Domagala, personal communication). In adults, whorl patterns are distinct only on the parietal scalp. Is the whorl pattern genetically controlled? A pair of homozygotic twins

were shown to have one and two whorls, respectively (Paine et al., 2004). Therefore, there must be an epigenetic component in hair whorl determination. While conserved molecular pathways underlie all hair follicle formation, local environmental and fortuitous factors can influence the final hair pattern.

In the mouse, transgenic mice that lose frizzled 6 show the formation of multiple whorls (Guo et al., 2004), suggesting the involvement of the Wnt pathway in this process. Interestingly, some strains of guinea pigs also show multiple whorls on their skin.

The formation of fingerprints is another example, as discussed in the next section.

### Periodic Patterning

This mechanism is most obvious in the formation of skin appendages and pigment patterns. During the formation of feather primordia, the epithelium has to become competent to respond to induction signals (forming a field). A reaction diffusion mechanism, involving activators and inhibitors, is proposed to operate in the dermal mesenchyme (Jung et al., 1998; Jiang et al., 1999). Through this mechanism, cells are set to become the primordia of skin appendages, stochastically. This then leads to the formation of the feather or hair primordia, evenly spaced with interfollicular skin. Similar processes were proposed for hair/wool formation (Nagorcka and Mooney, 1985; Moore et al., 1998; Meinhardt and Gierer, 2000). It should be emphasized that the process of periodic patterning can be uncoupled from the developmental wave process discussed in the previous paragraph.

The sequential appearance of feather buds is so exquisite (Fig. 3A) that it led scientists to propose models that are based on the use of previous buds as templates (Murray and Oster, 1984). The experiments by Jiang et al. (1999) showed that in a reconstitution situation, in which mesenchymal cells are dissociated into single cells, the periodic patterns will reform simul-

taneously. So the sequential appearance results from a global competence wave imposed on the local periodic patterning process.

Another dramatic example often referred to is the dissolution of pigment cells that leads to the formation of stripes on zebra fish or zebras and the formation of pigment ducts on fish or leopards. This was addressed earlier by Murray (1993). Recently, Prum and Williamson (2002) also applied a reaction diffusion mechanism to make a theoretical model of feather pigment patterns. However, some pigment patterns are controlled genetically by enhancer regions, as shown by differences in the Droopy Ear mouse mutant. Here, the normal, sharp delineation between dorsal and ventral pigmentation patterns is disrupted. This is produced by a loss of function mutation in TBox 15, which then allows agouti to be expressed further dorsally (Candille et al., 2004).

Do these patterns result from genetic coding or stochastic events? In fact, it is likely to be both. For example, consider patterns such as fingerprints (Kucken and Newell, 2005). They are used for individual identification because the ridge width and possibility for branching nodal points provide ample possibility for endless variation. Fingerprints among monozygotic twins have more similar attributes (similar width, organization plan) than with unrelated individuals, but they are nonidentical and are sufficiently different to be used as individual identifiers (Jain et al., 2002). Thus there is a nongenetic component at this level of tissue morphogenesis, where molecular codes become indirect and cells interact based on physical-chemical rules.

### Morphogenetic Gradient

We acknowledge the importance of molecular codes, and have proposed the skin Hox code hypothesis for regional specificity of skin and skin appendages (Chuong, 1993). Yet, how are these molecular codes set up?

A morphogenetic gradient, such as an Shh gradient, has been used

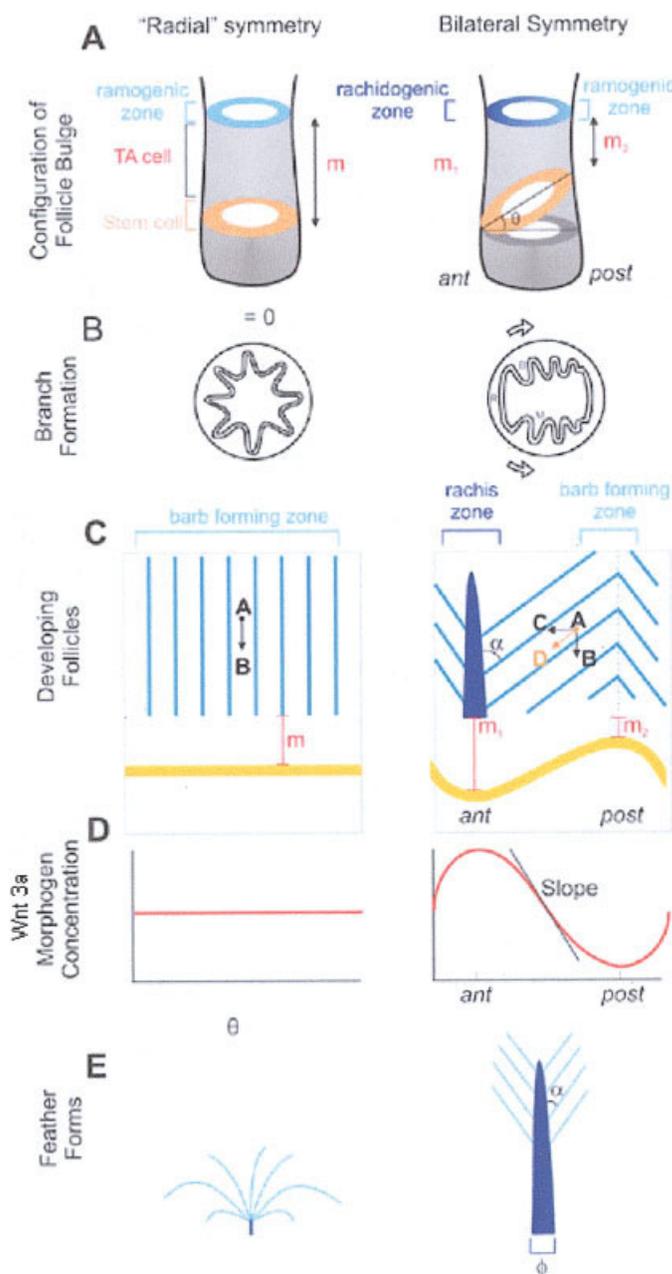
to explain dorsal-ventral spinal cord determination (Monsoro-Burq and Le Douarin, 1999) and anterior-posterior (A-P) limb bud patterning (McGlenn and Tabin, 2006). Here we will use a recent example describing how a Wnt 3a gradient in adult feather follicles patterns epithelial stem cells to form either radial or bilateral, symmetric feathers.

In the adult bird, there are radially symmetric body feathers and bilaterally symmetric flight feathers. Feathers do not contain the bulge structure found in hairs that house stem cells. We recently identified feather stem cells located as a concentric ring sitting at the bottom of the feather follicle (Yue et al., 2005). Interestingly, in radially symmetric feathers, the stem cell ring is placed horizontally. In bilaterally symmetric feathers, the stem cell ring tilts toward the anterior rachis position. This topological difference led us to propose that there would be a break of symmetry in the ramogenic plane where feather branches start to form (Fig. 4). Indeed, in bilaterally symmetric feathers, we found a Wnt 3a gradient from anterior to posterior positions that does not exist in radially symmetric feathers. Flattening the Wnt 3a gradient using RCAS retrovirus-mediated gene misexpression converted bilaterally symmetric feathers to radially symmetric feathers (Yue et al., 2006). A local Wnt 3a gradient released from a bead causes the forming barb ridge branches to swirl toward the Wnt 3a source. Swapping dermal papillae between radial and bilaterally symmetric feathers shows that the dermal papilla determines the gradient configuration and feather symmetry, while stem cells can respond to the morphogenetic gradient to make different forms of feathers. This is an excellent example, showing how a microgradient within a feather follicle can set the organ shape.

## PATTERNS THAT CAN BE CHANGED IN THE ADULT

### Traveling Wave

Hair follicles go through regenerative cycles: they cycle through growth



**Figure 4.** Morphogenetic gradient. Left column shows an idealized radially symmetric feather. Right column shows a bilaterally symmetric feather. **A,E:** The proximal follicle shows ordered compartments of stem cells (orange color), TA cells, and differentiating cells (ramogenic zone) (Yue et al., 2005). In radially symmetric feathers, the ring is horizontal. In the bilaterally symmetric feathers, the ring is tilted from zero to about  $45^\circ$ . The molecular gradient in the ramogenic zone is shown in shades of blue. **B,C:** In an open follicle preparation, the feather filament cylinder is opened to form a plane. In radially symmetric feathers, all new barb ridges form at the same time and in parallel. In bilaterally symmetric feathers, the tilting of the stem cell ring results in a discrepancy of maturation due to the fact that the TA cells have to travel (or are displaced) different distances before they reach the ramogenic zone ( $m_1$  and  $m_2$ ). On the anterior side, cells are more mature. The shift in cell position is represented by vectors AB, AC, and AD. **D:** According to this model, there should be a molecular gradient along the A-P axis. Indeed, we found a Wnt 3a gradient. Flattening the gradient converted feathers from bilateral to radial symmetry (Yue et al., 2006).

(anagen), regression (catagen), hair shaft shedding (exogen), and resting phases (telogen) (Paus and

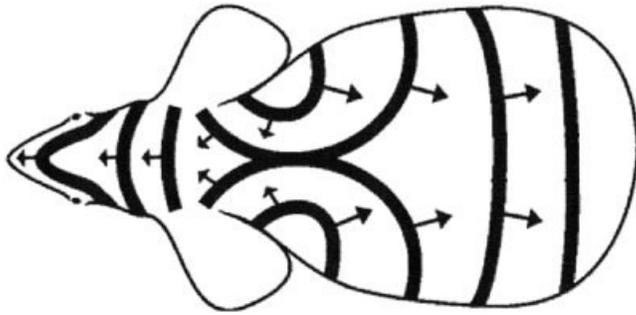
Cotsarelis, 1999; Stenn and Paus, 2001; Paus and Foitzik, 2004). Visualizing hair waves is facilitated by

hair cycle-dependent changes in integument pigmentation and timed hair loss in nude mice (Militzer, 2001; Suzuki et al., 2003), *Msx2* null mice (Ma et al., 2003; Mecklenburg et al., 2005), calcineurin B1 (Mammucari et al., 2005), etc. As a result, we observe skin regions cycling through discrete stages, each representing a different "status" of hair follicle (Fig. 1S and T). The regions can appear as waves of dramatic pattern changes across the adult mouse skin.

Militzer (2001) analyzed more than 400 nude mice on albino (NMRI, *foxn1<sup>nu</sup>*) and pigmented (C57BL/6, *foxn1<sup>nu</sup>*) backgrounds for more than one year. Pink skin turns dark when hairs enter anagen III and returns to pink when anagen is completed. Skin pigmentation changes travel in a wave-like fashion on the skin surface of these mice. When mice are young, all hair cycles initially synchronize, but with increasing age the hair cycles over different regions desynchronize. Thus, the skin pigment pattern breaks into distinct stripes and patches. As mice age, the stripes and patches become narrower/smaller and eventually appear random.

Dramatic traveling hair waves occur in the *Foxn1<sup>nu</sup>* strain of nude mice on the C57BL/6 background (Suzuki et al., 2003). These mutant mice have a distinct defect in the *Foxn1* gene that results in faster hair cycling. Thus the dynamic pigmentation pattern changes described above progress faster than those observed in classical nude mice. In young mice, the pigmentation oscillation takes place synchronously throughout the skin. The wider pigmented stripes progress to become narrower bands as a mice age. Some mice (usually more than seven months) show narrow, roughly evenly-spaced pigmented stripes that travel along the trunk; however, many mice show irregular, fragmented, or very wide stripes (Fig. 5).

The pattern can become more complex. Ma et al. (2003) reported "cyclic alopecia" in *Msx2* knockout mice. This phenotype is due to the fact that hair fibers are defective and are dislodged specifically during



**Figure 5.** Traveling stripes. **A:** In the adult mouse, hair follicles go through regenerative cycling. They appear as black in the anagen period. In this mutant nude mouse, hair filaments are lost in the telogen period and appear white. This helps us visualize the changing states of hair follicles, which appear as traveling waves (after Suzuki et al., 2003; Plikus and Chuong, 2004). Arrows describe the direction of wave propagation. **B:** *Msx2* null mice show cyclic alopecia in which hair shafts are dislodged at a specific time of hair cycle but can also regenerate. As a result, patches of hairy domains (black) and bald domains (white) are formed. These domains alternate between growth and resting phases, and give the impression of traveling stripes. The shape and size of these domains, their relative configuration changes over time, and situation in (A) is a special example of this phenomenon. Based on Ma et al. (2003).

the catagen phase. The skin of these mice during anagen is black and hairy, but during telogen is bald and nonpigmented. As the hairs cycle, the alopecic regions reenter anagen and regain pigmentation in a progressive order. Long-term observation of hairy and bald skin regions revealed a "cyclic alopecia" phenomenon. Hairs within one skin domain cycle in waves but not with hairs in neighboring domains (which also cycle in waves, but with an independent rhythm).

In essence, the "traveling stripes" of the *Foxn1<sup>nu</sup>* mice are a manifestation of the same phenomenon. Notch 1 activation in keratinocytes can go through the RBP or calcineurin B1 pathways. Recently, mice with a calcineurin B1 deletion also showed a cyclic alopecia phenotype (Mammucari et al., 2005). In adult humans, most hair follicle cycles independently, so there are no wave-like patterns.

### Hormone-Based Changes of Appendage Pattern

Since hair or feather follicles can go through regenerative cycles, a completely new type of skin appendage can reform with a new shape and size through a regenerative cycling mechanism. This is most obvious in sex hormone-dependent changes

(Mayer et al., 2004). Upon puberty, skin appendages in specific regions are transformed when sex hormone (estrogen and androgen) pathways are activated. Sex steroids can also affect the melanogenic activity of epidermal melanocytes, giving rise to hormonally-based changes of skin and skin appendage patterns as evident in birds (Fig. 2A).

This is most apparent in tail feathers of hens/roosters and peahens/peacocks. Sexual dimorphism characteristics are also observed in mammals, including humans (Wheeler, 1991). In the human beard, axilla, and genital regions, hair follicles are transformed from the vellus to the terminal state. With increasing age, the reverse tends to occur, leading to androgenic alopecia. Vellus hairs also can be transformed into unwanted terminal hairs (e.g., on the upper lip and lower legs) when properly stimulated by androgens, leading to hirsutism. Here, terminal hairs in the frontal and parietal scalp are affected, but not those in the occipital region. As a result, the type of hairs that form and the region of hair growth (hairline) change at different ages. The mechanism controlling how scalp and occipital hairs respond to sex hormones is not known, but appears to be mediated

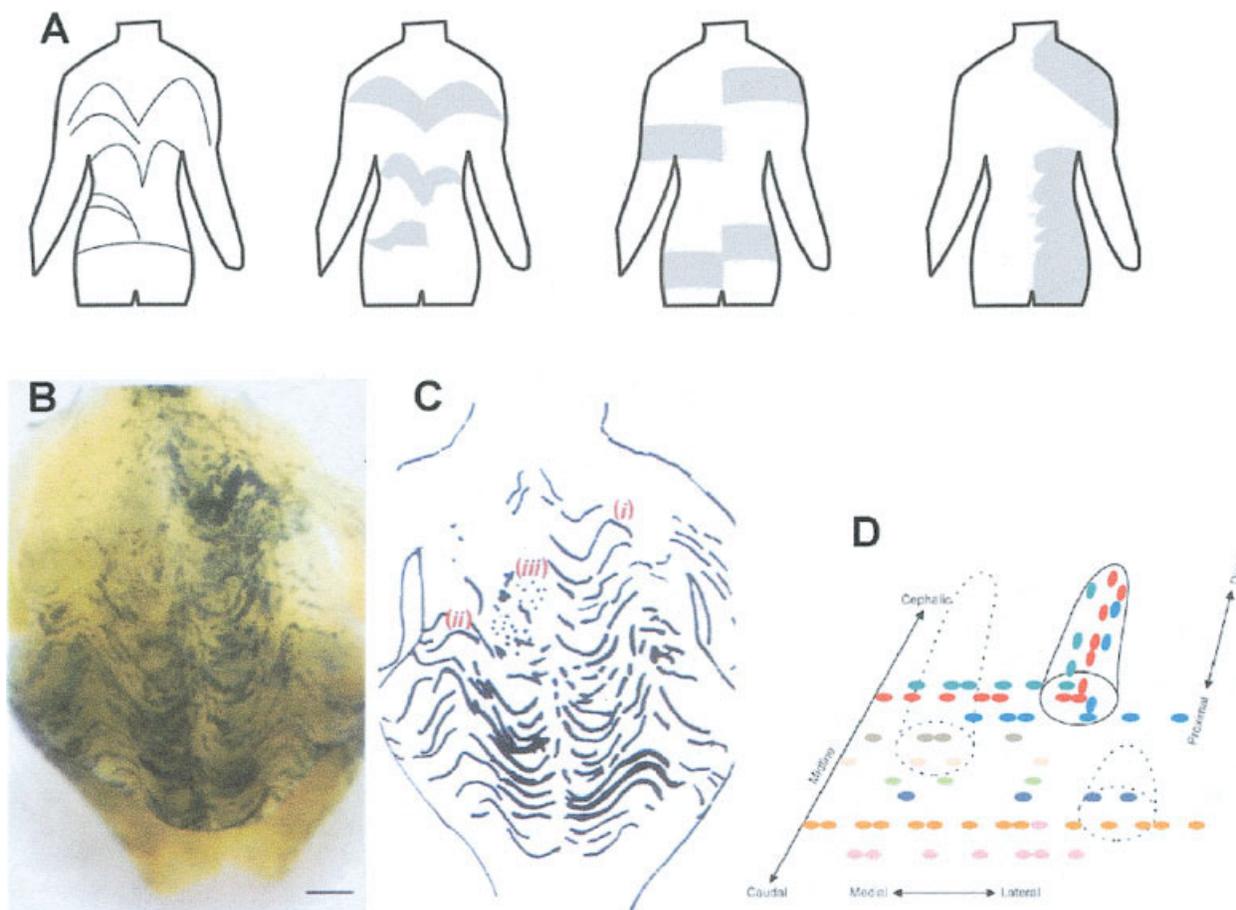
by differences in dermal papillae, which exhibit varying response to stimulation with androgens or estrogens (Randall et al., 2001; Inui et al., 2002; Conrad et al., 2005). In fact, estrogens and estrogen receptor-mediated signaling are powerful mediators or even inducers of wave pattern formation, namely of hair waves in mice (Ohnemus et al., 2006). Thus, hormonally-based skin lesion patterns are also the consequence of region-specific developmental programming.

### Environmentally-Based Pattern Changes

Change in light/dark cycles produced by the seasonal lengthening and shortening of days or changes in temperature can alter the type or coloring of skin appendages. This can be seen in the seasonal (summer/winter) hair coat variation of horses, snow shoe rabbits, etc. In nature, changes in the length of the light period are translated into changes in the plasma melatonin and prolactin levels, which can trigger animals to produce a longer/shorter or whiter/darker coat to improve their chances for survival during a given season (Rose et al., 1987). Now that we know that human and rodent skin and hair follicles are extrapituitary sites of melatonin synthesis (Sloiminski et al., 2002, 2003; Kobayashi et al., 2005), one wonders to what extent environmental cues (such as the length of the light period) can also affect seasonal changes in skin and skin appendage patterns.

### Pattern Formation of/in Skin Lesions

Since the skin covers the surface of an individual, patterns on the skin are the most recognizable. They have been used as diagnostic clues to the dermatologist (Ackerman, 1997; Bologna et al., 2003; Sterry et al., 2006). In addition to developmental and physiological causes, patterns on the skin that develop can be due to pathological or artifactual causes. A multiauthored review featuring several view points focusing on skin lesion patterns was recently edited by Dr. Ralf Paus for



**Figure 6.** Genetic mosaicism on the skin. **A:** Lines of Blaschko. Through X chromosome inactivation, the lineage of epithelia cells can be seen to be distributed in lines horizontal to the A-P axis. Several examples of checkerboard or patch patterns on human skin are seen in several human diseases (Happle, 1995, 2004). After Happle's viewpoint 2 in Chuong et al. (2006). **B:** Equivalent lines of Blaschko in embryonic chicken. Embryos are injected with nonreplicative virus carrying  $\beta$ -galactosidase. **C:** Line drawing of (B). **D:** Different cell lineages are represented by different colors. Analyses show that individual feather buds or individual barb ridges are made of cells from different lineages, not from a single lineage. Therefore, the local environment at the time of feather morphogenesis is more important than lineage. B-D: From Chuong et al. (1998).

*Experimental Dermatology* (Chuong et al., 2006b). Here, we will briefly summarize those discussions.

### Lineage, Genetically-Based

Molecular expression within cells is changed genetically or epigenetically during development. The changes can be transmitted to daughter cells because they involve somatic mutations in DNA or are mediated by epigenetic mechanisms such as X-chromosome inactivation, DNA methylation, etc. This collection of different patterns has mainly been studied in human diseases. The offspring of the mutated cells share a similar abnormality. The distinct phenotypes of these cells then manifest themselves

in the skin. These ectopic changes are named Naevus (Happle, 1995) (see Glossary). There are several striking examples in which lesions are limited to the left or right side of the body, regional segments, checkerboard patterns, or linear distributions (Fig. 6A) (Happle, 1993, 1995, 2004). The most striking example in the epidermal lineage is the Blaschko lines (Jackson, 1976). A recent case of linearly distributed acne turned out to be due to a somatic mutation in the FGF receptor in one epidermal cell lineage (Munro and Wilkie, 1998). The mechanism leading to the Blaschko lines is fundamental and not limited to humans. When early chicken embryo epidermal cells (embryonic day 2 [ED 2]) were la-

beled along the dorsal midline with replication-defective virus expressing  $\beta$ -galactosidase, their cellular descendents showed multiple parallel blue lines, resembling Christmas tree branches, radiating from the midline across the dorsal skin of late chicken embryos (Fig. 6B and C) (Chuong et al., 1998). However, analyses of these patterns show that formation of feather primordia or feather filaments are not based on lineage, but on the local environment at the time of formation (Fig. 6D).

There are many different types of ectodermal organs on the integument. Many of them share morphogenetic signaling pathways. Perturbation of one pathway can lead to changes in multiple organs (Plikus

et al., 2004). In humans, when a molecule, such as EDA, that is fundamental to these processes is mutated, it can lead to ectodermal dysplasia that affect multiple epithelial organs (Bologna et al., 2003).

### Anatomically- or Physiologically-Based

Distinct regions such as skin appendages, skin ridges, cutaneous nerves, blood vessels, etc., can contribute to patterns of skin lesions. When skin lesions develop, they may follow these obvious anatomical borders or follow "hidden" latent patterns based on physiological differences. Through various pathogenetic mechanisms, these different skin regions may result in different susceptibility to diseases. It is upon this dynamic landscape that skin lesions develop, and become distributed and shaped.

### Artificial

Human behaviors can also cause patterned lesions. For example, chronic sun exposure can lead to the characteristic ultraviolet (UV)-light-induced patterns corresponding to unclothed skin regions. Tattoos, skin paintings, hair dyes, cosmetic surgery, etc., can lead to further visible patterned changes on the skin.

### CONCLUSION

The skin is an excitable medium. In development, it conducts reactions among signaling molecules that determine the formation of skin appendages or the distribution of active melanocytes. In the adult, regenerative hair cycling provides a rich opportunity for the skin to renew itself based on hormonal and environmental changes. Patterns of skin lesions provide diagnostic clues to skin or systematic diseases. The convergence of genetic, epigenetic, and regenerative events to generate complex patterns on this very visible organ also provides a great experimental opportunity to study the many unknown mechanisms of biological pattern formation.

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