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REVIEW

The vertebrate tail: a gene playground for evolution

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Abstract
The tail of all vertebrates, regardless of size and anatomical detail, derive from a post-anal extension of the embryo known as the tail bud. Formation, growth and differentiation of this structure are closely associated with the activity of a group of cells that derive from the axial progenitors that build the spinal cord and the muscle-skeletal case of the trunk. Gdf11 activity switches the development of these progenitors from a trunk to a tail bud mode by changing the regulatory network that controls their growth and differentiation potential. Recent work in the mouse indicates that the tail bud regulatory network relies on the interconnected activities of the *Lin28*/*let-7* axis and the *Hox13* genes. As this network is likely to be conserved in other mammals, it is possible that the final length and anatomical composition of the adult tail result from the balance between the progenitor-promoting and -repressing activities provided by those genes. This balance might also determine the functional characteristics of the adult tail. Particularly relevant is its regeneration potential, intimately linked to the spinal cord. In mammals, known for their complete inability to regenerate the tail, the spinal cord is removed from the embryonic tail at late stages of development through a *Hox13*-dependent mechanism. In contrast, the tail of salamanders and lizards keep a functional spinal cord that actively guides the tail's regeneration process. I will argue that the distinct molecular networks controlling tail bud development provided a collection of readily accessible gene networks that were co-opted and combined during evolution either to end the active life of those progenitors or to make them generate the wide diversity of tail shapes and sizes observed among vertebrates.

Keywords Tail development · Tail regeneration · *Lin28* · *Hox* genes · Axial progenitors

Introduction

The tail is arguably among the most enigmatic parts of the vertebrate body. It is not a vital structure, yet it is fully conserved throughout vertebrate phylogeny. Indeed, even seemingly tailless animals contain a residual tail: the coccyx of humans or of tailless apes is one of the best-known examples. Another distinctive characteristic of the vertebrate tail is that, contrary to most body structures, we cannot assign a single function to it. Instead, the tail seems to be involved in a variety of different and sometimes remarkably unrelated roles [1], which is also reflected on the large diversity of sizes and structural features observed in the tails of different species. Many of those functions have a mechanical component. Improvement of locomotion performance is among the most prominent roles in this category. Particularly, animals living in aquatic environments, such as fish or crocodiles, use their tail mainly as a propelling device [2, 3]. Land vertebrates also benefit of the tail for locomotive purposes, either as an additional leg for kangaroos, facilitating walking mechanics in lizards, or serving as a counterbalance to help animals like cats or mice move through narrow or unstable surfaces [4–7]. An interesting variation upon this function was the evolution of prehensile tails, which allow grasping and holding to objects [8, 9]. Non-mechanical tail functions have also been described for some vertebrate species. For instance, in rats or beavers this structure has been shown to play important roles in body temperature regulation [10, 11], and the tail of the leopard gecko serves as a primary fat storage structure [12]. Also relevant is the tail's protective role in different vertebrates, both actively as a weapon [13] and passively by providing a large bodily target for predator attack that can be discarded without compromising animal's life, a process known as autotomy [14]. Finally, the

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tail serves as an important intraspecific communication tool [15].

What is then the origin of the tail and how has it evolved to produce such a wide diversity of anatomical and functional diversity observed across vertebrate phylogeny?

Back to basics: the tail bud and the embryonic development of the tail

The adult tail derives from a post-anal extension of the embryo typically referred to as the tail bud. The tail bud was first brought into the embryonic spotlight almost 100 years ago when Holmdahl proposed that vertebrate development occurs in two steps [16]. During the first step, the embryo would produce what he called the primary body (head and trunk), derived from the three germ layers and organized by the activity of the primitive streak. During the second step, the embryo would generate the secondary body (the tail), derived from of a blastema-like structure containing a homogeneous mass of mesenchymal cells, which we now know as tail bud [16]. Shortly thereafter, it was proposed that the tail bud was not a separate structure containing a somewhat homogeneous cell population, but was instead a mix of various cell types directly derived from the germ layers that form the more anterior embryonic regions [17, 18]. These rather conflicting views generated decades of discussion and have been more recently brought together as representing two sides of the same process [19]. However, notwithstanding the embryonic origin of the tail bud, the matter of its remarkable phylogenetic conservation is rarely addressed. This becomes especially relevant considering that, although its postembryonic product is clearly not essential for the animal's life, the tail bud is always present during embryogenesis, even in animals in which it does not contribute to any functionally relevant structure in the adult animal. In this review, I will suggest that the tail bud is the result of the mechanisms involved in the organized termination of the molecular and cellular activities that build the vital structures of the vertebrate body after their key function is complete. Those mechanisms would provide enough plasticity to the tail bud to allow evolutionary forces to generate tails of different shapes and sizes able to confer competitive advantages for the animal's survival in specific ecological niches.

The main driving force for body formation is a group of cells, collectively known as axial progenitors, located at the posterior embryonic end [20, 21]. These cells progressively lay down the anlage for the different body organs and structures as they extend the embryo posteriorly along its main body axis. The initial stages of post-cranial axial extension are dedicated to building vital organs. This process, which is the hallmark of trunk development, involves the activity of at least two major subsets of axial progenitors. One of them includes those generating the lateral and intermediate mesoderm that, together with the endoderm, form the organ systems involved in digestive, excretory and reproductive activities [21]. The second major group includes the progenitors for both the paraxial mesoderm, which forms the muscle-skeletal case supporting and protecting internal organs and the spinal cord that provides innervation to the different body structures [21]. The completion of trunk formation, roughly marking the end of Holmdahl's primary body, denotes the stage when the embryo contains the full complement of organs required to cover their vital and reproductive functions. After this point the axial progenitors producing the large catalog of tissues that compose trunk structures have also fulfilled their key developmental function and their activity should, therefore, be turned off. I will argue that it is the active process of progenitor termination what, in addition to building the structures that organize proper ending of trunk organ systems, also generates structures like the tail bud or the hind limb field and that these, in turn, supply a rich playground full of readily accessible gene networks that can be co-opted and combined either to end the active life of those progenitors, or to make them generate a variety of complex new structures.

The tail progenitors

Transplantation experiments demonstrated that the tail bud indeed contains cells with the functional features of axial progenitors [22–24]. Their origin and developmental potential were best revealed by retrospective cell tracing studies in the mouse. These showed that tail bud progenitors are direct descendants of a subset of those building trunk structures, most particularly the progenitors generating the neural tube, the paraxial mesoderm and the notochord, which incidentally represent major structural components of the embryonic tail [25]. These observations thus confirmed and refined the hypothesis of the continuity between trunk and tail structures initially proposed by Vogt and Pasteels [18].

The progenitors for the spinal cord and the paraxial mesoderm have attracted particular attention in recent years because, despite belonging to two different germ layers (ectoderm and mesoderm, respectively), a wealth of in vivo and in vitro data indicates that they both originate from a bipotent cell generally known as the neuro-mesodermal progenitor (NMP) [25–33]. Most studies involving these cells focused on their general properties and the mechanisms controlling their fate choices, and have been extensively reviewed elsewhere [34, 35].

However, recent data now indicate that, despite belonging to the same cell lineage and sharing many properties, trunk and tail bud NMPs actually represent distinct cell

populations. A key feature separating trunk and tail NMPs is the factors at the top of the regulatory hierarchy controlling their activity [36] (Fig. 1). Genetic experiments in the mouse indicate that trunk, but not tail, axial progenitors functionally require *Oct4* activity [37]. In particular, conditional *Oct4* inactivation at mouse embryonic stage (E) 7.5 (i.e. just before the beginning of extension through the trunk region) produced embryos lacking trunk structures [37], whereas inactivation at E9.0, shortly after the embryo has entered the tail bud stage, had no negative effect on axial extension [37]. In complementary experiments, it was shown that sustained *Oct4* activity in the progenitor region of mouse embryos both elongated the trunk and blocked tail bud formation [38]. The finding that the characteristically long trunk of snakes most likely results from sustained *Oct4* activity [38] further supports the notion that embryos need to down-regulate *Oct4* in the progenitor area to exit the trunk growth mode and enter the tail bud stage.

Oct4 activity (or in some species another homolog family member [39]) is not restricted to NMPs but is also essential for lateral and intermediate mesoderm progenitors to build trunk resident organ systems. The end of *Oct4* activity in axial progenitors thus marks the end of the organ-forming stage of development (Holmdahl's primary body). Two key questions arise then, both relevant to understand the origin of the tail bud: first, what are the mechanisms finishing the *Oct4*-dependent phase; and secondly, what is the fate of the progenitors after this stage. The answer to the former, which can be reformulated as the control of the trunk to tail transition, will surely be multilayered, as *Oct4* regulation is not uniform among vertebrates [38, 40]. Nevertheless, current data indicate a global role for Gdf11 signaling in this process. Indeed, it has been shown that in a wide variety of vertebrates, from amphibians to reptiles to mammals, *Gdf11* becomes activated at the axial level of the hind limb buds (which, as I will discuss later, acts as a proxy for the end of trunk structures) [38, 41], indicating that *Gdf11* activity might be a master regulator of the trunk to tail transition. Functional experiments support this idea. In the mouse, inactivation of *Gdf11* delays this transition (despite still happening due to partial redundancy with *Gdf8* [42]), resulting in a significant elongation of the trunk [43, 44]. This effect is not restricted to mammals, as in *Xenopus* embryos morpholino-mediated *Gdf11* down-regulation both increased trunk length and reduced tail bud size [45], and in chicken embryos molecular or chemical inhibition of this signaling also produced phenotypes compatible with trunk elongation [41, 46]. Conversely, complementary experiments involving premature activation of Gdf11 signaling produced significant anteriorization of the transition to tail development, with a dramatic reduction of the embryo's trunk size [38, 41, 44, 46]. The observation that increased *Oct4* activity in axial progenitors prevents their switch into tail development while forced Gdf11 signaling in these cells can override trunk formation indicates that Oct4 and Gdf11 are involved in a reciprocal negative functional regulation. However, the molecular details of this process remain to be clarified.

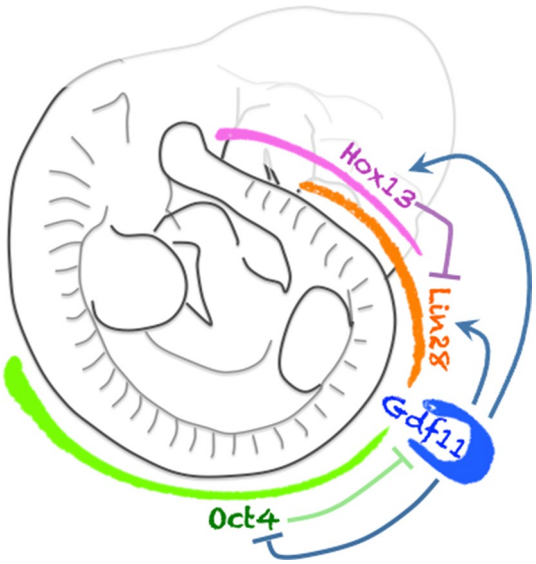

Fig. 1 Schematic representation of the basic regulators of trunk and tail development in mouse embryos. Oct4 is the main driver of trunk region development. At a given point, Gdf11 activity takes command and switches the regulatory processes from Oct4 to Lin28-dependent. Later in development, Gdf11 signaling promotes Hox13 gene expression, which counteracts Lin28 activity and finishes tail extension. Gdf11 and Oct4 activities are reciprocal negative regulators; the balance between these two activities determines the position where the embryo switches to tail bud development

Recent data identified some of the mechanisms by which Gdf11 activity triggers the trunk to tail transition. In the progenitors for the intermediate and lateral mesoderm, Gdf11 activates a program that organizes the end of excretory, genital and digestive systems by generating the mesodermal components of the cloaca, while concomitantly inducing the termination of lateral plate mesoderm through the formation of the hind limb buds [44]. In contrast, NMPs activate a regulatory network different from that operating in the organ-building progenitors [44], one that will ultimately lead to the formation of the tail bud. In particular, recent reports revealed that Gdf11 signaling brings NMPs under the control of a regulatory system that includes the *Lin28*/*let-7* axis and *Hox* genes of the paralog group 13 as key players [47, 48]. *Lin28* genes encode RNA binding proteins that have been involved in the control of stem cell activity [49, 50] and their relevant role in tail bud progenitors was shown by gain and loss of function experiments in the mouse. In particular, inactivation of *Lin28a* produced significant tail shortening, whereas over stimulating *Lin28* gene expression increased the number of tail vertebrae [47, 48] (Fig. 2). Further genetic studies

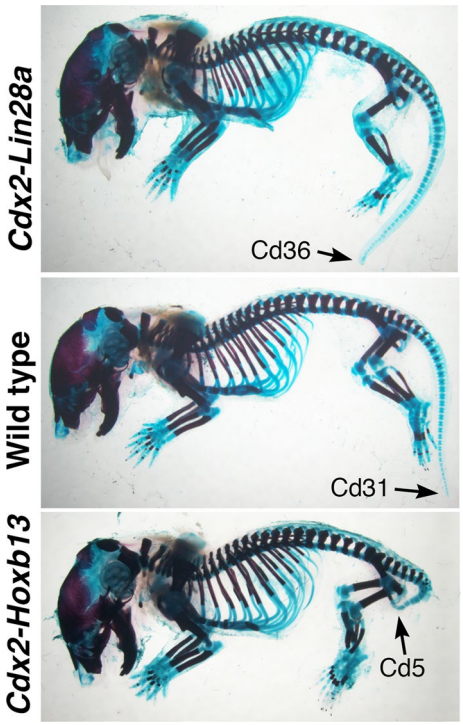

Fig. 2 Skeletal phenotype of mouse newborns with increased *Lin28a* or *Hoxb13* expression. The middle animal shows a wild type control, containing 31 caudal vertebrae (Cd). The upper animal shows a transgenic animal expressing *Lin28a* in the axial progenitors, increasing the number of caudal vertebrae to 36. The lower animal corresponds to a transgenic animal with increased *Hoxb13* expression in the axial progenitors, reducing the number of caudal vertebrae to 5

also indicated that *Lin28* controls the activity of tail bud axial progenitors by blocking maturation, and thus activity, of the *let-7* miRNA family, which is also a major Lin28 target in a variety of biological systems [50]. Indeed, *let-7* family loss and gain of function experiments in the mouse produced phenotypes opposite to those resulting from equivalent experiments with *Lin28*: *let-7* overexpression decreased tail length, while reduction of *let-7* activity significantly increased tail vertebral counts [48]. The finding that simultaneous activation of both *let-7* and *Lin28* genes reproduced the *let-7* overexpression phenotype (tail shortening) further supported that *Lin28* activity is mediated by *let-7* inactivation [48].

Hox genes of paralog group 13 also play a fundamental role in the control of tail bud NMP activity. In mouse embryos, *Hoxb13* and *Hoxc13* are activated in the progenitor region of the tail bud relatively late in development, particularly when compared to their A and D cluster counterparts, at the time when tail extension starts to decline [47, 51, 52]. But perhaps more importantly, experimental modulation of their expression impacts tail growth. In particular, *Hoxb13* mutant mice have additional tail vertebrae [51] and premature activation of either *Hoxb13* or *Hoxc13* in the axial progenitors using a transgenic approach produces a variable reduction in tail size [47, 53] (Fig. 2).

A common characteristic of the phenotypes resulting from gain or loss of *Lin28*, *let-7* or *Hox13* gene expression is that they are restricted to the tail bud axial progenitors and their derivatives, with little or no effect on trunk NMPs irrespective of expression levels [47, 48]. These observations further highlight the existence of intrinsic functional differences between trunk and tail NMPs, despite the latter being direct derivatives of the former according to cell tracing experiments [25]. These differences are further supported by in vitro culture experiments. It has been shown that the capacity of the progenitor area of mouse embryos to produce colonies when cultured in vitro is progressively reduced as development proceeds, mirroring the decay in *Oct4* expression [54]. Consistent with this, explanted cells from the progenitor region at the tail bud stage—hence lacking *Oct4* activity-, survive in culture only for short periods of time and fail to double their population more than four times [47, 54]. Inhibition of Gdf11 signaling, however, rescues the capacity of tail bud cells to be cultured long-term in vitro, somehow mimicking the increase in tail bud progenitors observed in *Gdf11* mutant embryos in vivo [47]. Importantly, this rescue is independent of *Oct4*, relying instead on *Lin28* genes. Also, these cell culture experiments indicate that *Hox13* genes most likely play a significant role in the decay of tail bud progenitor lifespan by interfering with *Lin28* expression. Therefore, the balance between *Lin28* and *Hox13* activities seem to be at the core of the network regulating the survival ability of tail bud NMPs, at least in mouse embryos.

Functional differences between trunk and tail bud NMPs are also conveyed into their neural and mesodermal derivatives. The neural tube is one such case, where these dissimilarities were first recognized decades ago. In particular, the spinal cord is built by primary neurulation during trunk development, whereas tail neural tube is made by secondary neurulation [55]. Comparable functional differences have also been observed in cells entering mesodermal fates: while there is no apparent discontinuity between somitogenesis in trunk and tail regions, they are differentially affected by specific genetic modifications. In particular, blocking *Lfng* cycling activity or reducing its expression levels interfere with trunk but not tail somite formation [56, 57]. Conversely, forced expression of *Hoxb6* in the presomitic mesoderm blocks tail bud but not trunk somitogenesis [58]. Thus, an ever-growing body of functional and molecular data indicates that, despite the morphological continuity of the anterior–posterior axis supported by common regulatory proteins (e.g. Brachyury, Wnt3a and Fgf8 [34, 35]), trunk and tail progenitors are initiated by different genetic programs facilitating species-specific tail innovation.

Connecting tail bud NMP regulatory network with cell metabolism

It has been recently reported that tail bud NMPs rely on aerobic glycolysis instead of oxidative respiration [59]. Accordingly, inactivation of the glycolytic pathway in chicken embryos resulted in axial truncations. Interestingly, glucose metabolism is one of the main processes under the control of the *Lin28/let-7* axis [60]. These observations hint at the possibility that these genes might regulate tail bud NMP activity by controlling the metabolic pathways operating in these cells. So far, this hypothesis has not been directly tested. Yet, gene expression data from *Gdf11* mutants, which have increased *Lin28* expression, are consistent with this possibility, as the progenitor-containing region of *Gdf11* mutant embryos showed enrichment in the expression of genes coding for key enzymes involved in aerobic glycolysis [47].

Interestingly, blocking aerobic glycolysis in chicken embryos down-regulated Wnt and Fgf signaling, as well as *T (Brachyury)* expression in the tail bud [59], thus suggesting that the control of metabolic pathways might be the functional link between *Lin28/let-7* and the expression of global regulators of NMP activity [34, 35]. Whether or not this hypothesis is indeed correct will require direct experimental evaluation.

The tail neural tube

Consistent with the known developmental potential of NMPs, the embryonic tail extends by generating a neural tube flanked by somite pairs throughout its whole length. However, not all vertebrates have the whole set of derivatives expected from these embryonic precursors in their adult tail. In particular, the mammalian tail is mostly composed of vertebrae and muscles (somite derivatives), with no associated spinal cord for most of its length. In the mouse, only the first 4 or 5 from the nearly 31 caudal vertebrae enclose spinal cord [47, 51]. This is likely a general mammalian characteristic, as similar observations have been reported for the spider monkey, the rat and the cat. This feature is also reflected in the anatomy of caudal vertebrae described in other mammalian species, which consistently show the absence of spinal cord canal after a precise axial level in the tail [61–65]. Conversely, the tail of non-mammalian vertebrates, including crocodiles, lizards and salamanders, contain a fully developed spinal cord [66–68]. Motor and sensitive innervation of tail structures in lizards and salamanders depend on the associated spinal cord [67, 68]. However, in mammals, tail innervation is provided by neurons located at sacral and anterior caudal levels, indicating that there seems to be no need for a tail central nervous system to coordinate complex tail movements [61, 62, 64]. Even in the newly grown tail of lizards after autotomy, innervation of the tissues in the tail regenerate is provided by neurons in the stump of the sectioned spinal cord and not from the regenerated neural tube [67], thus further calling into question the actual need for a spinal cord in the adult tail.

Interestingly, tails containing or lacking spinal cord have clearly different regeneration potential (Fig. 3). Indeed, the mammalian tail totally lacks such potential, whereas lizards and salamanders effectively regrow their tails and a series of resection and transplantation experiments have clearly established a fundamental role for the spinal cord in tail regeneration processes [69, 70]. It has been shown that radial glial cells within the ependymal canal are the key elements in this regeneration activity [71–73]. These cells delaminate from the tip of the sectioned spinal cord, invade the blastema formed at the injury site and rebuild a new spinal cord that eventually organizes the whole tail regeneration process.

The central role of the spinal cord in tail regeneration is illustrated by the anatomy of the tail regenerate. In particular, while in salamanders the newly grown tail is identical to the original [68, 74], the tail regenerate in lizards is structurally different, being essentially composed of a non-segmented cartilage rod enclosing the neural tube, surrounded

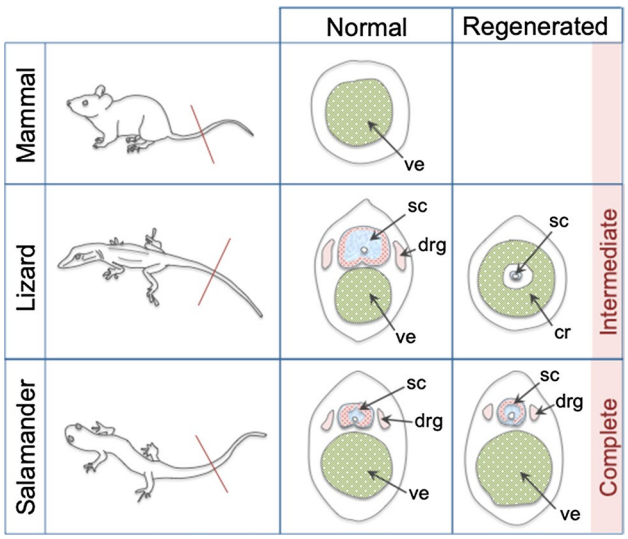

Fig. 3 The neural tube is a key regulator of vertebrate tail regeneration. Mammals, whose tails do not have neural tube, are unable to regenerate their tails. Lizard tails have intermediate regeneration properties. Their tail spinal cord grows but fails to reproduce normal patterns, leading to the formation of a tail regenerate made out of a cartilage rod (cr) enclosing the spinal cord (sc), but no individual vertebrae (ve). Salamander tail regeneration is complete, reproducing normal tail anatomy, with individual vertebrae, well-patterned spinal cord and peripheral neural structures such as dorsal root ganglia (drg)

by muscle masses formed by long instead of short muscle fibers [67, 74] (Fig. 3). Importantly, these differences correlate with the characteristics of the spinal cord in their tail regenerate. In salamanders, the new structure keeps normal features, including robust dorso-ventral patterning and the re-establishment of a properly organized peripheral nervous system, including dorsal root ganglia [71]. On the contrary, the spinal cord in the lizard tail regenerate is dominated by ventral Shh signals, lacking normal dorsal components [74]. These observations indicate that similarly to what occurs during embryonic development [75], morphogenesis of the regenerated vertebral elements is guided by signals from the neural tube acting on adjacent mesoderm, with a complete set of dorsal and ventral activities operating in salamanders and just ventral signals in lizards. A variety of grafting and signal mimicking experiments provided experimental support to this hypothesis [74].

While it seems clear that the neural tube is regenerated from their ependymal radial glial cells, the origin of the cells that rebuild mesodermal tail structures is less well defined. Experiments in different animal models indicate that the origin of these tissues varies among species. It has been reported that in axolotls radial glial cells at the tip of the sectioned spinal cord delaminate from the neural tissue to invade the surrounding blastemal mesenchyme, and contribute to both skeletal and muscle structures in the tail regenerate [71, 73], thus suggesting a common origin for neural and mesodermal tissues in their regrown tail. However, in other species mesodermal tissues in the tail regenerate seem to derive from mesodermal tissues instead of from spinal cord cells [76, 77]. These observations suggest a correlation between the degree of accuracy in the tail regeneration process and the plasticity of the ependymal radial glial cells. Indeed, the observation that these cells seem to regain a differentiation potential resembling that of embryonic NMPs suggests that in salamanders the radial glial cells maintain a high degree of plasticity, allowing them to revert to a more undifferentiated state to engage in a regeneration process that closely resembles embryonic tail growth. On the contrary, adult tail regeneration in lizards would be different from embryonic tail growth, relying on the partial plasticity of radial glial cells to regenerate incomplete neural structures, and on muscle and cartilage cells from the tail stump to produce new mesodermal tissues [76].

Single cell analyses of the regenerating *Xenopus* tadpole tail have identified a new cell type, known as regeneration-organizing cell (ROC), that plays an essential role during both development and regeneration this structure [78]. ROCs can be identified in the epithelium covering the developing tadpole tail. Upon injury, these cells move to cover the amputation plane, where they function as an organizing center that signals to promote the proliferation of progenitors for the different tissues involved in tadpole tail regeneration [77, 78]. This mobilization requires activity of reactive oxygen species (ROS), which have been shown to be produced by the wound and to play essential roles in tadpole tail regeneration [78–80]. Interestingly, signaling through the TGFβ pathway is an essential component of ROC signaling activity, thus connecting this regeneration process to normal tadpole tail development [45, 81]. It should be noted that the *Xenopus* tadpole represents a late embryonic stage in some animal's life and thus its regeneration might be different from that observed in tails of adult animals. It will be important to understand whether ROCs or ROC-like cell types are also involved in regeneration processes such as those occurring in the tails of adult salamanders or lizards.

The absence of neural tube might be the key to the lack of regeneration potential of the mammalian adult tail. Studies performed in mice indicate that neural tube elimination from the mammalian embryonic tail is an active process involving apoptosis [51, 82] and suggest a functional connection with the process halting tail bud NMP activity. In particular, *Gdf11* also seems to be a key regulator of this process. Indeed, newborn *Gdf11* mutant embryos contain a fully developed neural tube until the caudal end of their body axis, indicating that it was not resorbed from the tail bud at late developmental stages [47]. Moreover, tail bud cells that grow in vitro in the absence of Gdf11 signaling preferentially take neural fates after several days in culture [47], further supporting a negative effect of Gdf11 on neural differentiation from tail bud NMPs. It is possible that *Lin28* genes play a role in this process, as it is up regulated both in the neural tube of *Gdf11* mutant embryos and in cultured tail bud cells undergoing neural differentiation under Gdf11-blocking conditions [47]. The finding that *Lin28* genes promote neural stem cell growth [49] is consistent with this hypothesis. This idea is, however, not free of controversy as it has been described that in mouse embryos overexpressing *Lin28* or reducing *let-7* activity in tail bud progenitors makes them preferentially take mesodermal fates [48]. The origin of the differences between the two studies is unclear and might only be solved upon identification of the mechanisms of *Lin28* activity in these progenitors.

Additional data from mouse embryos indicate that, in addition to their activity on tail bud progenitors, *Hoxb13* and *Hoxc13* could also play a fundamental role downstream of Gdf11 in actively promoting tail neural tube removal [47]. These genes are strongly expressed in the region of the tail neural tube fated to elimination from the adult tail [47, 51, 52]. In addition, the spinal cord of *Hoxb13* and *Hoxc13* mutant mice extends farther caudally into the adult tail than in wild type mice [51, 52], indicating that tail neural tube removal is incomplete in these mutants. Redundancy between these two genes might explain the absence of a more pervasive phenotype of spinal cord persistence in each of the single mutant mice. This possibility has so far not

been tested directly, but the co-occurrence in *Gdf11* mutant embryos of a fully developed spinal cord and complete absence of *Hoxb13* and *Hoxc13* expression is consistent with this hypothesis [47].

On the basis of the aforementioned role that the spinal cord plays during lizard and salamander tail regeneration, together with the molecular characteristics of the tail bud spinal cord and the timing of its removal from the mouse embryonic tail, I would suggest that the presence of neural tube in the tail of mammalian embryos might be required to guide proper differentiation of adjacent somites to build the appropriate surrounding vertebral elements. Yet, after this function is complete, tail neural tube becomes dispensable and is, therefore, eliminated as part of normal development.

A genetic playground for evolution

The data discussed in this review indicates that the tail bud results from a change in the gene regulatory program controlling NMPs, coincident with the end of the trunk forming stage of embryonic development. Most of what is currently known about the genetic activities involved in this process derives from experiments performed with mouse embryos, but it is possible that other vertebrates share at least some of the components identified in this animal model. Clearly, Gdf11 activity seems to act as the master switch controlling the change from trunk to tail developmental modes in most vertebrates [41], and it is likely that the *Lin28/let-7/Hox13* network also operates at least in the tail bud of other mammals. Temporal, spatial or quantitative variations in the opposite activities provided by these (and maybe other yet to be discovered) factors resulting either from gene expression levels, intrinsic activities of their gene products, or timing of their activation, could impact strongly on the growth and differentiation properties of the tail bud. Evolutionary forces could thus select for variants in these parameters, providing a functional balance that would result in tail phenotypes advantageous for specific ecological niches. In human embryos, for instance, premature activation of *Hox13* genes would be enough to promote a fast switch off of tail bud NMP activity and produce just a residual tail, represented by the coccyx, that seems to be favorable for bipedalism [83]. Premature *Hoxb13* activation might also be in the origin of the absent tail in chicken, as this gene seems to be expressed at a rather early stage of tail bud development (geisha.arizona.edu).

Although there is still no data indicating whether the same gene network operates in lizards or salamanders, it can still be used to illustrate how the complex tails of these animals could derive from variations in the activity of the tail bud regulatory network. For instance, the size and anatomical features of the lizard and salamander tail are compatible with absent *Hox13* activity (as they are long and contain a fully-grown neural tube). As these genes are indeed expressed in their tail buds [84–86] the apparent lack of response to *Hox13* genes could be explained if, contrary to what is observed in mice, Gdf11 did not render NMPs competent to respond to *Hox13* activity. This type of scenario would resemble the inability of snake and Paenungulata embryos to respond to the rib blocking activity of Hox10 proteins, resulting from a polymorphism in an enhancer that mediates Hox10 functional input [87]. Variable changes in activities inhibiting NMP function could also be the basis for the different levels of plasticity left in cells derived from tail bud progenitors that eventually drive the tail regeneration capabilities of salamanders and lizards.

In conclusion, irrespective of how similar gene networks regulating tail bud NMP activity turn out to be in the different vertebrates, a general principle can still be postulated, according to which Gdf11 signaling activates a tail-specific gene regulation program in NMPs. While integrated to some extent with the trunk-extending molecular network, the existence of specific factors at the top of the genetic hierarchy controlling progenitor activity in the tail bud might have provided a fertile playground on which evolutionary forces could modify the tail with limited effects on other embryonic regions. This way, selecting for the combination of gene activities that resulted in particular, optimally adapted, tail phenotypes to each particular environment would have thus generated the rich tail diversity observed among vertebrates.

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