Development 138, 2401-2415 (2011) doi:10.1242/dev.040972

An eye on the head: the development and evolution of craniofacial muscles

Ramkumar Sambasivan[1,2,*], Shigeru Kuratani[3,*] and Shahragim Tajbakhsh[1,2,*,†]

Summary

Skeletal muscles exert diverse functions, enabling both crushing with great force and movement with exquisite precision. A remarkably distinct repertoire of genes and ontological features characterise this tissue, and recent evidence has shown that skeletal muscles of the head, the craniofacial muscles, are evolutionarily, morphologically and molecularly distinct from those of the trunk. Here, we review the molecular basis of craniofacial muscle development and discuss how this process is different to trunk and limb muscle development. Through evolutionary comparisons of primitive chordates (such as amphioxus) and jawless vertebrates (such as lampreys) with jawed vertebrates, we also provide some clues as to how this dichotomy arose.

Key words: Craniofacial muscle development, Cranial mesoderm, Genetic regulation, Head muscle evolution, Tbx, Pitx

Introduction

The skeletal muscles of the head, known as craniofacial muscles (Fig. 1), are essential for everyday movements, including those that control facial expression, mastication and eye movements. Myofibres are the functional unit of all skeletal muscles and have a common contractile function, yet a number of observations have highlighted that an extraordinary diversity exists among different muscles, including between the groups of craniofacial muscles. Such diversity at the molecular level, in terms of the expression of specific metabolic activities or contractile protein isoforms (Cheng et al., 2004; Fischer et al., 2002), is likely, but not proven, to underlie the adaptation of these muscle groups to the varied functional tasks they perform. Significantly, such diversity could be a potential basis, at least partly, for the differential involvement of muscle groups in various myopathies (see Glossary, Box 1). For example, many myopathies cause severe dysfunction of some skeletal muscles, but, intriguingly, other muscles are functionally spared. How does this diversity arise? Do the developmental and genetic programmes of skeletal muscles impinge on their differentiated phenotype and hence contribute to this diversity?

Traditionally, studies of developmental myogenesis have focussed on trunk and limb musculature; thus, less is known about the development of head muscles. However, recent studies are beginning to shed light on the mechanisms that regulate craniofacial muscle development. Notably, embryological and genetic studies have shown that different strategies establish myogenesis in different regions of the organism during development. A striking difference between cranial and trunk myogenesis, for example, is that the trunk paraxial mesoderm (see Glossary, Box 1), an embryonic tissue that generates trunk and limb muscles, is segmented into transient epithelial somites (Fig. 1), whereas the cranial mesoderm (CM, see Glossary, Box 1), which is the source of most head muscle progenitors and was

Box 1. Glossary

Branchiomeric muscles. A term used for pharyngeal (visceral) arch-derived muscles.
Cardiomyogenic progenitors. Precursors of heart muscle.
Chondrichthyans. Fish with a cartilaginous skeleton, such as sharks and rays.
Cranial lateral mesoderm. Lateral portion of the cranial mesoderm close to the pharynx (also called the pharyngeal mesoderm or cranial splanchnic mesoderm) that gives rise to the anterior heart field and most first and second pharyngeal arch-derived muscles.
Cranial neural crest (CNC). CNC arises from the dorsal portion of the anterior neural tube and populates the face and the pharyngeal arches. It gives rise to bones, cartilage, nerves and connective tissue, and part of the base and facial bones of the skull.
Cranial paraxial (paraxial head) mesoderm. Bilateral mesenchymal mesoderm parallel and immediately adjacent to the neural tube/notochord; it generates a subset of extra-ocular, and other head, muscles.
Cucullaris muscles. A group of neck muscles connecting the skull to the shoulder girdle.
Cyclostomes. Greek for 'round mouth'. Refers to modern jawless vertebrates including lampreys and hagfishes.
Epibranchial placodes. Cranial ectodermal thickenings that contribute to cranial ganglia, particularly cranial nerves VII, IX and X.
Epidermal placodes. Epidermal thickenings that give rise to sensory organs and cranial ganglia.
Head cavities. Cranial mesoderm organised as epithelial cysts in some vertebrates.
Hypobranchial muscles. A group of neck muscles located in the floor of the pharyngeal arches.
Myopathies. A group of muscle-wasting diseases.
Outflow tract. Ventricular portion of the heart through which blood passes in order to enter the great arteries.
Pharyngeal arches. Literally meaning a gill-supporting structure, present in all vertebrate embryos and give rise to structures of the face, ear and neck.
Prechordal mesoderm. Vertebrate-specific (also known as axial) mesoderm found anterior to the notochord and ventral to the neural tube. It is displaced bilaterally during development and contributes to certain extra-ocular muscles in chick.
Trunk paraxial mesoderm. Mesoderm that lies parallel to and either side of the trunk embryonic body axis. It segments into epithelial somites that harbour dermis, skeletal muscle, brown fat, smooth muscle, bone and endothelial progenitors.

[1]Institut Pasteur, Stem Cells and Development, Paris, F-75015, France. [2]CNRS URA 2578, 25 rue du Dr Roux, Paris, F-75015, France. [3]Laboratory for Evolutionary Morphology, RIKEN Center for Developmental Biology, 2-2-3 Minatojima-minami, Chuo-ku, Kobe, Hyogo 650-0047, Japan.

*These authors contributed equally to this work
†Author for correspondence (shaht@pasteur.fr)

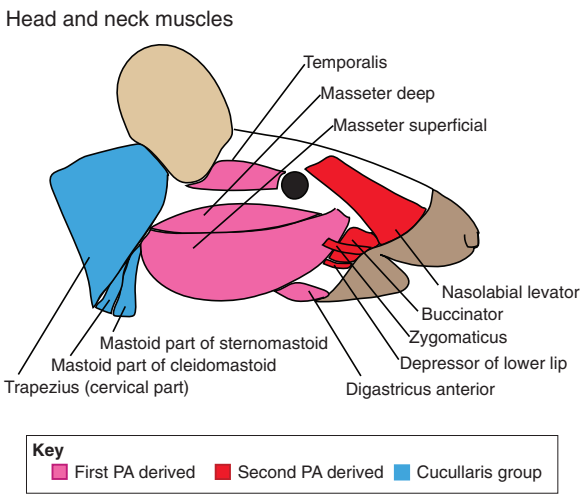

Fig. 1. Head and neck muscles. (**A**) Schematic of the vertebrate head muscles discussed in this review, showing the different facial muscles (red), which arise from the second pharyngeal arch (PA), and jaw muscles (pink), which derive from the first PA. Also shown are the neck muscles (the cucullaris group, blue), which we discuss in this review because they develop in the transition zone from trunk to head. (**B**) Detailed view of the extra-ocular muscles showing the following muscles: the inferior rectus (IR); medial rectus (MR); superior oblique (SO); levator palpebrae (LP); superior rectus (SR); lateral rectus (LR); and retractor bulbi (RB). Reproduced with permission from Sambasivan et al. (Sambasivan et al., 2009). The muscles shown all derive from the cranial mesoderm. Muscles in the head that arise from trunk mesoderm, such as the tongue muscles, are not referred to as head muscles in this article.

historically considered to be morphologically segmented, does not appear to be overtly segmented in vertebrates (Wedin, 1949; Kuratani, 2008b; Kuratani et al., 1999). Indeed, the organisation of the CM is currently delineated by molecular markers rather than by overt anatomical boundaries (Bothe and Dietrich, 2006). Another peculiarity of cranial myogenesis is its link to heart development; the CM generates cardiomyogenic progenitors (see Glossary, Box 1) as well as craniofacial muscles. This is an issue of considerable interest, as it provides clues to understanding the evolution of the precursor tissue of head muscles. Other muscles in, or associated with, the head, such as tongue and some neck muscles, are derived from muscle founder cells located in the anterior-most somites. These cells migrate distally before differentiating at their final location. In general, neck muscle development is less well studied, but it has also been considered here in an evolutionary and genetic context as it provides additional clues to the modular vertebrate design.

In evolutionary terms, the head of vertebrates is thought to be a novel structure (Gans and Northcutt, 1983; Northcutt and Gans, 1983) and some head muscles are also a vertebrate novelty. The major part of the head evolved from a non-vertebrate chordate ancestor with its anteroposterior (AP) positional landmarks preserved (Yu et al., 2007). Simultaneously, new cellular components, such as cranial neural crest and epidermal placodes (see Glossary, Box 1) evolved as additions to the 'real head' during this evolutionary transition from protochordates to vertebrates (Gans and Northcutt, 1983; Northcutt and Gans, 1983). Therefore, muscles might also have originated multiple times as independent events (Gans and Northcutt, 1983) and, consequently, head muscles could have arisen independently of trunk muscles.

The evolutionarily novel vertebrate head muscles, which include extra-ocular muscles (EOMs), jaw muscles and facial muscles (Fig. 1), are what we refer to collectively as craniofacial muscles and are the focus of this review. They all derive from the CM, an embryonic tissue that is also unique to vertebrates, the evolutionary origins of which are still unclear. This review first highlights our current understanding of the organisation of the CM, focussing on new insights into the molecular basis of craniofacial development and how this process is different to trunk and limb muscle development. With the help of recent reports on the genetics of head muscle development, notably that distinct gene networks govern the onset of cranial myogenesis, we also discuss the evolution of craniofacial muscles. We then discuss the development and evolution of muscles in the neck, in light of the possible dichotomy (between the trunk and cranial mesoderm) of their origin. Finally, we discuss the link between cardiac muscle fate and craniofacial muscle development, a link that sets craniofacial muscle development apart from trunk muscle development.

Cranial mesoderm organisation

To understand craniofacial muscle development, the issue of how the CM is organised first needs to be discussed. Most muscles of the head derive from the CM, which also gives rise to other cell types (Fig. 2), including cardiomyocytes (Kelly et al., 2001; Mjaatvedt et al., 2001; Waldo et al., 2001), the posterior part of the neurocranium (the portion of the skull that encases the brain), and angiogenic cells (Couly et al., 1992; Couly et al., 1993; Evans and Noden, 2006; Hacker and Guthrie, 1998; Noden, 1983b). Fate-mapping studies in quail-chick chimeras and lineage-tracing studies that have used dye or retroviral labelling in chick (Couly et al., 1992; Couly et al., 1993; Evans and Noden, 2006; Hacker and Guthrie, 1998; Noden, 1983b) and mouse embryos (Gage et al., 2005; Trainor and Tam, 1995) have identified regions of the CM that are myogenic. Some of these studies identified specific regions of the CM as being a source of founder cells for distinct muscle groups in the head (see Noden and Francis-West, 2006).

Developmental history of cranial mesoderm

One potential source of confusion about the CM is its lack of clear anatomical boundaries or defined molecular markers to distinguish between the different mesoderm populations in the head. By contrast, somitic and lateral plate mesoderm (LPM) in the trunk are delineated morphologically, as well as by gene expression profiles, as distinct physical entities (Goulding et al., 1994; Palmeirim et al., 1997; Tajbakhsh and Buckingham, 2000) (Fig. 2). At an open neural plate stage [Hamburger-Hamilton stage (HH)7-8 (1-3 somites) (Hamburger and Hamilton, 1992) in chick, around embryonic day (E)8.0 in mouse], position and boundaries set by other tissues, as well as marker gene

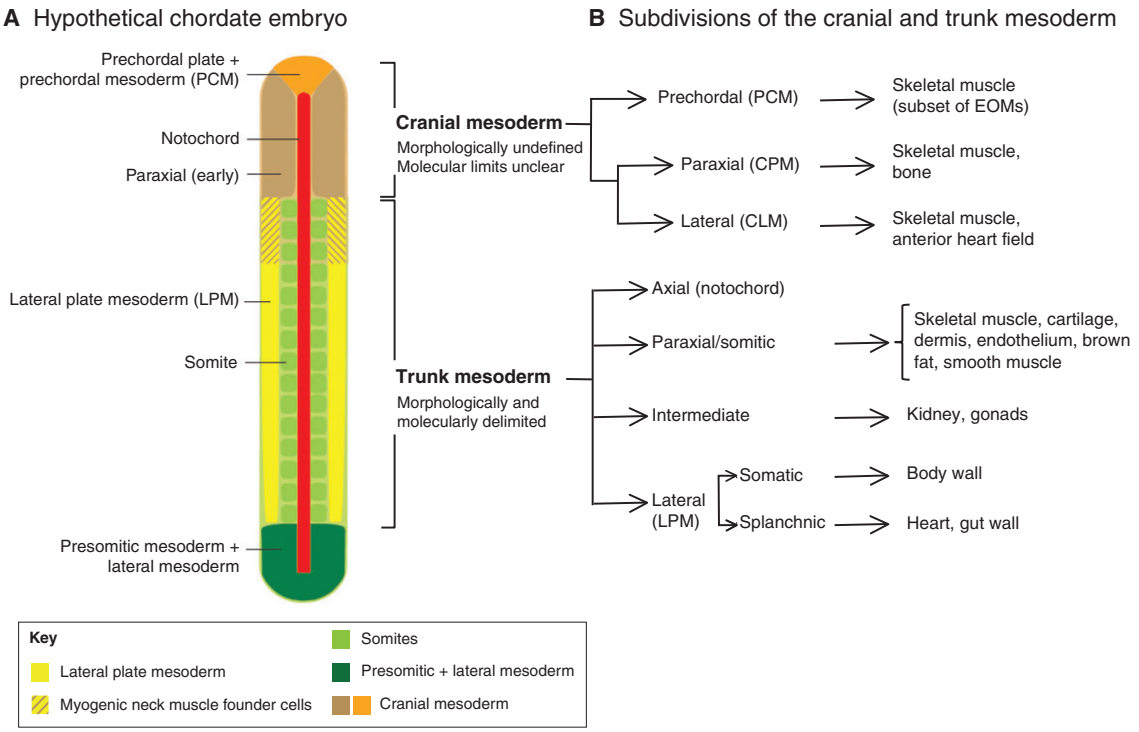

Fig. 2. Subdivisions of the cranial mesoderm. (**A**) Schematic of a hypothetical, 15-somite chordate embryo, viewed dorsally, showing the tissues that give rise to the trunk and cranial musculature, lateral plate mesoderm (LPM, yellow); somites of the trunk mesoderm (pale green); and the presomitic and lateral mesoderm (dark green), which give rise to the somites and LPM, respectively. The yellow and brown hatched LPM denotes the presence of myogenic neck muscle founder cells. Other tissues shown include the prechordal plate and prechordal mesoderm (orange) and the notochord (red). (**B**) Subdivisions of the cranial and trunk mesoderm and the descendents they give rise to (right). Cranial mesoderm, which sits parallel to the notochord in early embryos (~3-somite stage), is divided into prechordal mesoderm (PCM), cranial paraxial mesoderm (CPM) and cranial lateral mesoderm (CLM), based on fate mapping and gene expression data. Trunk mesoderm is divided into axial, paraxial, intermediate and lateral plate mesoderm (LPM).

expression, delineate the CM into prechordal, cranial paraxial and cranial lateral mesoderm (see Glossary, Box 1). When the neural tube closes dorsally and the endoderm ventrally (HH8-10 in chick and E8.5-9.0 in mouse), the position and boundaries of the tissues alter. Prechordal mesoderm (PCM, see Glossary, Box 1; Fig. 2), which develops in an axial position beneath the neural plate and is located anterior to the notochord, is subsequently displaced caudally and laterally during development (accompanying the lateral growth of the anterior neural plate). It then integrates with the remaining CM, which is located anterior to the somites (Fig. 2). Thus, the CM at an early stage does not include the PCM, yet at later stages it does. Furthermore, the initial mediolateral organisation of the CM is largely transformed into a dorsoventral pattern. Moreover, for the mesoderm above and below the pharynx, a new mediolateral divide emerges by means of marker gene expression and cell fate choices. Where a particular cell population is located, and at which stage of development this localisation occurs, is yet to be established. Later, when pharyngeal arches (PAs, see Glossary, Box 1) form, a new system of organisation is established.

The mediolateral and AP compartmentalisation of the CM, in terms of fate, has been proposed based on studies of avian embryos. Lineage tracing studies have shown that the medial-posterior region of the CM is destined to contribute to part of the neurocranium and the medial-anterior region to the extrinsic eye muscles, whereas the lateral region contributes to the PA derivatives, including the craniofacial muscles (Couly et al., 1992; Evans and Noden, 2006; Noden and Francis-West, 2006). These studies also showed that the PCM is displaced bilaterally during development and that it contributes to a subset of EOMs. However, the extent of this contribution and its requirement for the development of this set of muscles is not known. The restricted expression of genes such as *Pitx2*, *Alx4*, *Msc*, *Twist* and *Tbx1* transcription factors (see below) reveal that molecular differences exist along the mediolateral and AP axes of the CM (Bothe and Dietrich, 2006).

CM organisation: evolutionary considerations

Although the CM does not appear to be partitioned morphologically, an obvious question is whether or not the CM is regionalised. Regionalisation of the CM into 'medial' and 'lateral' components has been previously suggested (Couly et al., 1992; Evans and Noden, 2006), and previous interpretations of comparative embryology have divided the CM along the AP axis into head cavities (see Glossary, Box 1) (see Kuratani, 2003). The CM has also been divided into dorsal and ventral compartments, representing the somatic (paraxial) and visceral (pharyngeal arch) mesoderm as cephalic counterparts of somites and lateral plate, respectively (Goodrich, 1930; Jarvik, 1980; van Wijhe, 1882).

Whether the CM is segmented has been extensively debated [for discussion of both sides of the argument, see reviews by Wedin (Wedin, 1949), Holland (Holland et al., 2008) and Kuratani (Kuratani, 2008b)]. Views on the segmentation of the vertebrate

head were stimulated by the discovery of head cavities in the late nineteenth and early twentieth centuries. A comparative zoological view is that segments or somites in bilaterians originate from reiterating mesodermal epithelial cysts, or myogenic coeloms. Thus, the evolutionary origin of the vertebrate head mesoderm has been dealt with in the same context. One typical theory of head mesoderm evolution proposes that the three pairs of coeloms found in deuterostome larvae originated from four gut diverticula (outpouchings) in cnidarians, with the posterior pair of cavities being secondarily subdivided to generate somites (Starck, 1978). Thus, the vertebrate paraxial mesoderm was classified into three groups: premandibular mesoderm, more posterior head mesoderm, and somites.

In some basal gnathostomes (jawed vertebrates), the CM is organised into a successive series of these epithelial cysts (Balfour, 1878; Goodrich, 1930; Kuratani, 2005; van Wijhe, 1882). In many chondrichthyans (see Glossary, Box 1), three pairs of cavities are observed and are called, from anterior to posterior, the premandibular, mandibular and hyoid cavities; they are ventrally associated with PA mesoderm. Of these, the premandibular cavity was assumed to generate four of the EOMs innervated by the oculomotor nerve, the mandibular cavity contributes to superior oblique muscle innervated by the trochlear nerve, and the hyoid cavity contributes to the lateral rectus innervated by the abducens. Thus, each head cavity is thought to produce EOMs, based on histological observations. In other gnathostome lineages (such as amniotes), head cavities are absent and EOMs appear to arise from mesenchymal CM, as well as from the prechordal plate (Couly et al., 1992; Jacob et al., 1984; Noden et al., 1999; Wachtler et al., 1984). Currently, divergent views persist regarding the evolution of head cavities: (1) head cavities are homologous to some rostral somites in the amphioxus, a group of non-vertebrate chordates (Beaster-Jones et al., 2006; Holland, 2000); (2) head cavities are serially homologous to the somites of the trunk (Jacobson, 1988); or (3) head cavities are entirely different from somites, and have a secondary origin in the lineage of either gnathostomes or vertebrates (Kuratani, 2003). To complicate matters further, the presence of head cavities in the lamprey, a jawless vertebrate, is itself contested (Wedin, 1949; Beaster-Jones et al., 2006; Holland et al., 2008; Kuratani, 2003; Kuratani, 2008b; Kuratani et al., 1999; Neal, 1918).

Studies of gastrulating chick embryos provide another view on the issue of CM segmentation. Here, the apparently unsegmented CM experiences two pulses of expression of the Notch pathway segmentation clock gene, *hairy1* (Dubrulle and Pourquie, 2002; Jouve et al., 2002). In the PSM, each pulse of this gene corresponds directly to the formation of a somite (Maroto and Pourquie, 2001). This finding indicates that: (1) the CM is not, as suggested previously (Jacobson, 1988; Jacobson, 1993), segmented into six cephalic somitomeres that are equivalent to somites, given that only two pulses of *hairy1* are observed (Jouve et al., 2002); (2) the PCM and the rest of the CM represent two segments, even though the PCM is not initially bilateral and is considered by some to be axial mesoderm owing to its initial position (Seifert et al., 1993); or, (3) the cyclic expression of *hairy1* in the CM does not define segments in the same fashion as it does in the PSM (Dubrulle and Pourquie, 2002).

Regionalisation based on anatomical landmarks

One way of assessing regional differences in the CM, especially along the AP axis, is to consider adjacent anatomical landmarks, such as the neural tube (Fig. 3). The organisation of the CM becomes more apparent when domains of cranial neural crest are also taken into account. Neural crest cells and the cranial nerves that arise from the neural tube harbour positional information along the AP axis (Fig. 3A,B). In addition, a close interaction occurs between neural crest cells and the mesodermal progenitors of the PAs and EOMs, with the neural crest playing a critical role in skeletal muscle cell migration, displacement and patterning (Ericsson et al., 2004; Grammatopoulos et al., 2000; Köntges and Lumsden, 1996; Matt et al., 2008; Noden, 1983a; Noden and Trainor, 2005; Olsson et al., 2001; Rinon et al., 2007; Schilling and Kimmel, 1997). Although muscle cell fate is not overtly affected when neural crest cells are genetically perturbed, a role for neural crest cells in regulating these muscle stem cells has not been ruled out (Rinon et al., 2007; von Scheven et al., 2006a).

The position-based associations between the cranial nerves (CN) and skeletal muscles are most evident between the so-called branchiomeric nerves (the nerves associated with the PAs, called CN V, VII, IX and X) and arch-derived muscles (Fig. 3C). The patterning of these nerves is guided by neural crest cells and epibranchial placodes (see Glossary, Box, 1), which are distributed in a specific fashion with respect to the segmentation of the PAs. This results in a typical topographical association between the nerves and the arch-derived muscles (Cordes, 2001). Moreover, it is the crest-derived ectomesenchymal cells in the PAs that provide the connective tissues to these muscles (Couly et al., 1993; Evans and Noden, 2006; Köntges and Lumsden, 1996; Noden and Francis-West, 2006). Thus, it is generally agreed that regional differences exist in the CM, but the link between the molecular profile and the future cell fate of the derivatives of CM is unknown. Furthermore, the initial overlap in the expression of skeletal myogenic and cardiomyogenic markers in CM has made it difficult to demarcate boundaries in this mesoderm (Grifone et al., 2007; Tzahor, 2009).

In essence, there is a lack of apparent morphological partitioning of the CM on any of the axes, thus distinguishing it from the somitic trunk mesoderm. Therefore, at least two possibilities can be suggested for the origin of the embryonic tissue that generates the evolutionarily recent craniofacial muscles. First, the CM could be a modified form of the visceral mesoderm (which arises from primitive gut) (see Kelly, 2010). This supposition agrees with the hypothesis that the CM is segmented in the form of the head cavities as entities that are comparable to somites. This is because the head cavities, which were reported to exist in early vertebrates, are equated, by comparative morphology, to the anterior somites of amphioxus, which also arise from the primitive gut (see Holland et al., 2008). A second possibility is that, from an evolutionary perspective, the CM is a novel tissue.

Genetic networks distinguish cranial and somitic myogenesis

Head muscles are the only musculature to develop in mice carrying mutations in certain myogenic genes, whereas other mutations cause severe developmental abnormalities in head muscles but not in trunk muscles (Table 1). Furthermore, recent findings show that distinct genetic regulatory cascades operate within individual craniofacial muscle groups, the EOMs and the PA-derived jaw and facial muscles. This highlights several interesting points that might reflect on putative subdivisions within the CM. Below, we discuss findings from mouse genetic studies that have revealed divergence in the craniofacial and trunk muscle developmental programmes.

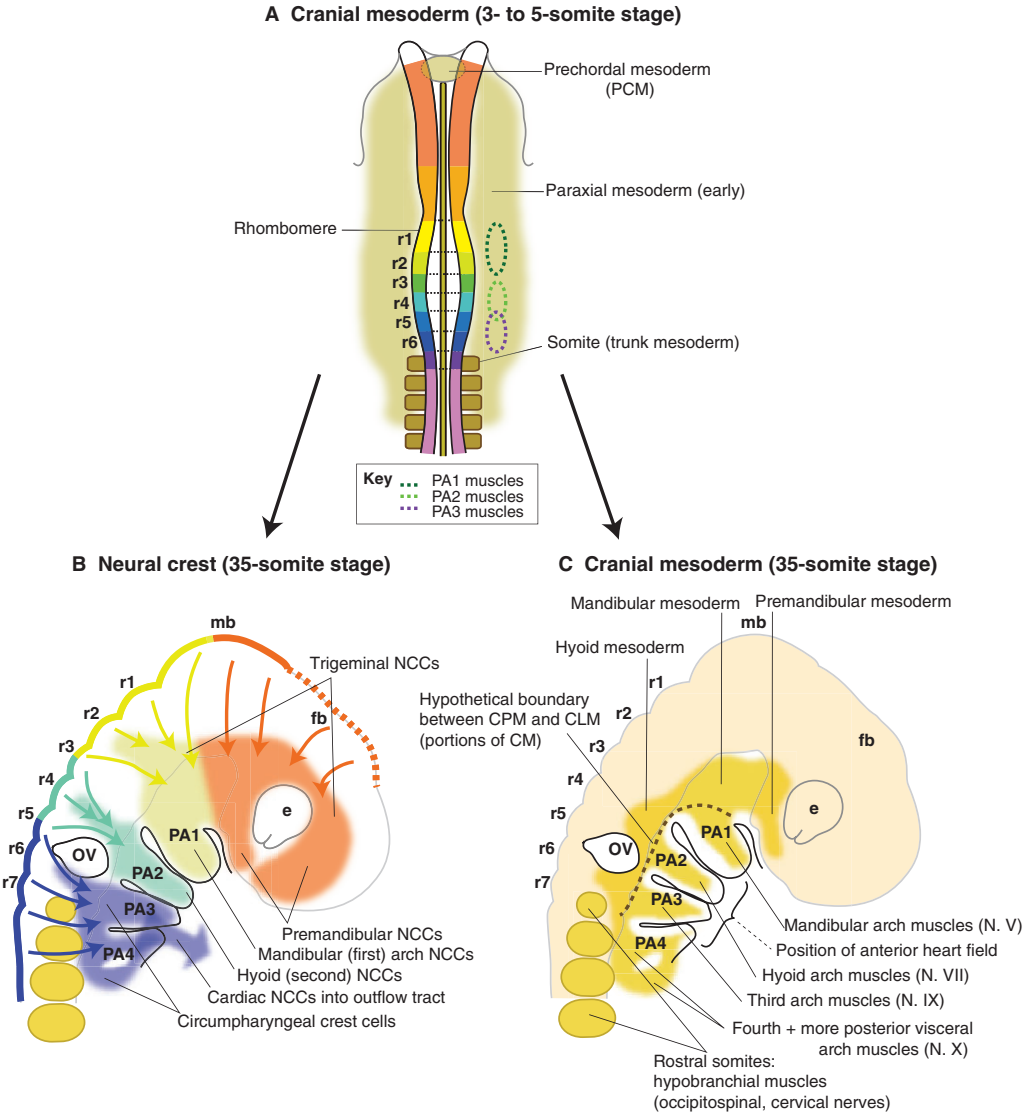

Fig. 3. Organisation of the cranial mesoderm relative to somites and neural crest. The cranial mesoderm (CM) lacks morphological boundaries but its organisation becomes more apparent when it is compared to domains of cranial neural crest. (**A**) Schematic of a 3- to 5-somite chick embryo in dorsal view, showing rhombomeres (r)1-6 (highlighted in yellow, green and blue), from which the neural crest originates (Creuzet et al., 2002). Rostral is top. The rhombomeres, the midbrain (light orange) and the forebrain (dark orange) regions that generate neural crest are indicated. The light green, dark green and purple dotted circles indicate regions of the CM that contribute to the different muscles of the pharyngeal arches (PA): PA1, PA2 and PA3, respectively. Image modified with permission from Creuzet et al. (Creuzet et al., 2002). (**B**) A lateral view of a 35-somite stage chick embryo, rostral uppermost, dorsal left, showing neural crest migratory routes to PA1-4. The rhombomeres (r), midbrain (mb) and forebrain (fb) regions are depicted in the same colours used in A (Creuzet et al., 2002; Kulesa and Fraser, 2000). (**C**) Schematic as in B showing maturing CM (yellow), which gives rise to premandibular and PA-derived muscles, as well as to founders of the anterior heart field. The PAs confer a new level of organisation to CM; at this stage, the CM in the arches is called PA mesoderm. The specific pattern of nerve supply (indicated in brackets) to muscle groups derived from each PA further highlights CM organisation at this stage. The position of the anterior heart field, which generates cardiomyocytes, is indicated. Images in B and C modified with permission from Creuzet et al. (Creuzet et al., 2002), and Hacker and Guthrie (Hacker and Guthrie, 1998). CLM, cranial lateral mesoderm; CPM, cranial paraxial mesoderm; e, eye primordium; fb, forebrain; mb, midbrain; NCCs, neural crest cells; N.III–N. XI, cranial nerves; ov, otic vesicle; PA1-PA4, pharyngeal arches; r1-r7, rhombomeres.

The myogenic regulatory factors

The core myogenic network consists of the basic helix-loop-helix (bHLH) myogenic regulatory factors [MRFs; *Myf5*, *Mrf4* (*Myf6* – Mouse Genome Informatics), *Myod* (*Myod1* – Mouse Genome Informatics) and *myogenin*], which determine muscle identity and/or promote muscle differentiation (Sambasivan and Tajbakhsh, 2007; Weintraub et al., 1991). These factors have been ‘redeployed’ during evolution to build the new head muscles. However, their roles differ in EOM versus PA skeletal muscle founder cells. For example, in the mouse, EOM development depends on either *Myf5* or *Mrf4*, and EOMs require both for robust myogenesis; interestingly, in their absence, *Myod* expression is severely compromised in EOM progenitors (Sambasivan et al., 2009). This is also the case for the epaxial muscles in the tail, which are missing in *Myf5:Mrf4* double mutant mice (Kassar-Duchossoy et al., 2004). Intriguingly, although *Myf5* plays a critical role in regulating PA muscle progenitors as well (Sambasivan et al., 2009), the genetic hierarchy upstream of the MRFs that specify PA and EOM muscle progenitors is distinct.

Although MRFs act as a gateway into myogenesis in all muscles, there are intriguing combinations of individual MRFs that function in different muscle groups within different vertebrates. In the mouse, *Myf5* and *Mrf4* act genetically upstream of *Myod* in EOMs, as they do in the somite-derived muscle groups. In zebrafish, *myf5* has been reported to be required for head muscle development (Chen and Tsai, 2002; Lin et al., 2006). However, in another zebrafish study, *myod* (*myod1* – Zebrafish Information Network), but not *myf5*, was proposed to be necessary for cranial myogenesis (Hinits et al., 2009). This is a notable inter-species difference, given that *Myf5* is the key player in cranial myogenesis in mouse (Sambasivan et al., 2009). As for *Mrf4*, in the absence of Myf5 and Myod, it drives robust differentiation of mouse

Table 1. Null mutants that affect head, trunk/neck and limb muscle myogenesis

Transcription factor	Family/expression/role	Head muscle	Trunk/neck muscle	Limb muscle	References
Pitx2*	**Family: Homeodomain.** Expressed in various embryonic tissues. Role in L/R asymmetry and specification of cell fates.	***Pitx2* null:** EOM progenitors mislocalized†; deficit of 1st PA-derived muscles.	***Pitx2* null:** Hypaxial myotome and body wall muscles deformed/absent†.	***Pitx2* null:** Limb muscle anlagen deformed†.	(Gage et al., 1999; L'Honoré et al., 2010; Kitamura et al., 1999; Shih et al., 2007)
Tbx1*	**Family: T box.** Expressed early in development. Speculated to have a role in regional specification of distinct subset of cell populations‡.	***Tbx1* null:** Random unilateral loss of 1st PA muscles†; loss of 2nd PA muscles†. ***Tbx1:Myf5* double null:** Almost complete loss of 1st PA muscles; loss of 2nd PA muscles.	***Tbx1* null:** Loss of cucullaris neck muscles and muscles of larynx†. ***Tbx1:Myf5* double null:** No apparent synergy on trunk myogenesis.	***Tbx1* null:** No apparent limb muscle deficiency. ***Tbx1:Myf5* double null:** No apparent synergy on limb myogenesis.	(Kelly et al., 2004; ‡Papaioannou and Silver, 1998; Sambasivan et al., 2009; Theis et al., 2010)
Msc, Tcf21*	**Family: bHLH.** Thought to be transcriptional repressors that delay commitment to muscle lineage.	***Msc:Tcf21* double null:** No temporalis, masseter and pterygoid muscles (1st PA subset); other muscles of 1st PA are present. All zebrafish cranial muscles lost in loss of functions.	***Msc:Tcf21* double null:** No apparent trunk musculature deficit.	***Msc:Tcf21* double null:** No limb musculature deficit reported.	(Lee et al., 2002; Lu et al., 2002)
Pax7*	**Family: Paired box.** TF upstream of MRFs; also expressed in CNC and developing CNS.	***Pax7* null:** Loss of postnatal muscle stem/progenitor cells; N/A in adult.	***Pax7* null:** Loss of postnatal muscle stem/progenitor cells; not required in adult.	***Pax7* null:** Loss of postnatal muscle stem/progenitor cells; not required in adult.	(Kuang et al., 2006; Lepper et al., 2009; Oustanina et al., 2004; Relaix et al., 2006; Seale et al., 2000) (§R.S. and S.T., unpublished)
Pax3	**Family: Paired box.** TF upstream of MRFs; also expressed in CNC and developing CNS.	***Pax3* null:** Head muscles present. ***Pax3:Myf5 (Mrf4)* null:** Head muscles present; EOMs missing as in *Myf5:Mrf4* double null; no synergy.	***Pax3* null:** Deficits in trunk, diaphragm and tongue muscles. ***Pax3:Myf5 (Mrf4)* null:** Missing trunk muscles.	***Pax3* null:** Limb muscles missing. ***Pax3:Myf5 (Mrf4)* null:** Limb muscles missing.	(Bober et al., 1994; Franz et al., 1993; Goulding et al., 1994; Tajbakhsh et al., 1997)
Six1, Six4	**Family: Homeodomain.** Expressed in various embryonic tissues. Specify cell fates.	N/A	***Six1:Six4* double null:** Severe trunk muscle deficit; no limb muscles.	***Six1:Six4* double null:** Limb muscles missing.	(Grifone et al., 2005)
Eya1, Eya2	**Family: Eya domain.** Expressed in various embryonic tissues. Specify cell fates.	N/A	***Eya1:Eya2* double null:** Absence of hypaxial somite derived muscles.	***Eya1:Eya2* double null:** Limb muscles missing.	(Grifone et al., 2007)
Meox1, Meox2	**Family: Homeodomain.** Expressed in various somite-derived embryonic tissues. Specify cell fates.	N/A	***Meox1:Meox2* double null:** Deficient trunk myogenesis.	***Meox1:Meox2* double null:** Deficiencies in limb muscles.	(Mankoo et al., 2003)
Myf5, Mrf4, Myod	**Family: bHLH.** Muscle cell fate determination factors; Mrf4 and Myod also regulate differentiation.	***Myf5 (Mrf4):Myod* null:** No myoblasts or differentiated cells throughout embryo. ***Myf5:Myod* null:** Failure of head muscles; some Desmin+ myoblasts via Mrf4 in EOM and PAs. ***Myf5:Mrf4* null:** EOM development fails.	***Myf5 (Mrf4):Myod* null:** No myogenesis in the embryo. ***Myf5:Myod* null:** Failure of post-embryonic trunk myogenesis. ***Myf5:Mrf4* null:** Myogenesis proceeds following a 1- to 2-day delay; epaxial muscle defects.	***Myf5 (Mrf4):Myod* null:** No myogenesis in the embryo. ***Myf5:Myod* null:** Failure of post-embryonic limb myogenesis. ***Myf5:Mrf4* null:** No significant limb muscle phenotype.	(Kassar-Duchossoy et al., 2004; Rudnicki et al., 1993; Tajbakhsh et al., 1997) (R.S. and S.T., unpublished)
Myogenin	**Family: bHLH.** Muscle differentiation factor.	N/A	***Myogenin* null:** Severe deficiency in prenatal but not adult muscle differentiation.	***Myogenin* null:** Severe deficiency in prenatal but not adult muscle differentiation.	(Hasty et al., 1993; Knapp et al., 2006; Meadows et al., 2008; Nabeshima et al., 1993)

bHLH, basic helix-loop-helix; CNC, cranial neural crest; CNS, central nervous system; EOM, extra-ocular muscles; L/R, left/right; MRF, myogenic regulatory factor; N/A, data not available; PA, pharyngeal arch; TF, transcription factor.
*Expression not restricted to muscle progenitors.
†Possible non-cell autonomous role.

embryonic somitic progenitors, but in this double mutant it fails to support the differentiation of EOMs or first PA-derived muscles (Kassar-Duchossoy et al., 2004). This could be due to differences in the somitic and cranial myogenic cell populations, a different threshold level of an MRF needed to initiate myogenesis, or the lower potency of Mrf4 as a transcription factor compared with Myod and Myf5 (Bergstrom and Tapscott, 2001; Weintraub et al., 1991). In zebrafish somites, Mrf4 does not support myogenesis when *myf5* and *myod* are compromised, but it has the potential to do so when supplied ectopically (Hinits et al., 2009; Schnapp et al., 2009). However, Mrf4 is not expressed during early head muscle development (Hinits et al., 2009).

These differences in MRF functions across phylogeny imply that these transcription factors have acquired new roles during evolution. The ancestral MRF gene is presumed to have given rise to the four family members in vertebrates (Atchley et al., 1994). *Myf5* and *Mrf4* are genetically linked and separated by 9 kb, suggesting that this is the most recent duplication event. In addition, there is compelling evidence to suggest that the functions of each of the MRFs are evolutionarily conserved. For example, *Xenopus* and human Myf5 are highly conserved outside the bHLH region, yet human Myf5 and Myod are divergent outside the bHLH region. This suggests that, throughout evolution, the MRF paralogues have diverged functionally whereas the orthologues have not drifted significantly. In this context, why different MRFs are used in different locations in the head, as is seen in epaxial and hypaxial trunk myogenesis (Tajbakhsh and Buckingham, 2000), remains unclear. Analysis of the combinatorial factors acting on enhancer sequences of each MRF for different organisms should provide further insights into this issue.

Pax3 and Pax7

In somitic muscle founders, *Pax3* and *Pax7*, paired-homeodomain genes, act genetically upstream of *Myod* (Tajbakhsh et al., 1997), and they are likely to activate *Myod* expression by binding to its regulatory sequences (Hu et al., 2008). *Pax3* expression is absent in head muscle stem/progenitors (Hacker and Guthrie, 1998; Sambasivan et al., 2009; Tajbakhsh et al., 1997). Furthermore, in *Pax3:Myf5:(Mrf4)* mutant embryos (in which embryonic *Mrf4* expression is perturbed in cis by disruption of the *Myf5* locus) trunk muscles fail to form whereas most head muscles develop (Tajbakhsh et al., 1997).

Pitx2 and Tbx1

Pituitary homeobox 2 (Pitx2) is a bicoid-related homeodomain transcription factor (Gage et al., 1999; Kitamura et al., 1999) expressed widely in the head of mouse embryos, including within the CM and the pharyngeal ectoderm. It plays a key role in specifying EOMs, which are missing in *Pitx2*-null mice, and it is a key regulator of PA muscle development, (Gage et al., 1999; Kitamura et al., 1999; Shih et al., 2007). *Pitx2* is expressed in trunk muscle progenitors, but it is not required for embryonic trunk myogenesis (Kitamura et al., 1999). Furthermore, *Pitx2* has been shown to act downstream of *Pax3* in somitic myogenesis (L'Honore et al., 2010). *Pitx2* directly binds the promoters of *Myf5* and *Myod*, and mice that lack *Pitx2* in the mesoderm that gives rise to EOMs fail to develop these muscles (Zacharias et al., 2011). However, *Pitx2* does not activate *Myod* expression in *Myf5:Mrf4* double mutants, which lack EOMs, in spite of its continued expression in mutant EOM founder cells (Sambasivan et al., 2009), although it does so in limb muscle progenitors (L'Honore et al., 2010) suggesting that other co-factors play critical roles with *Pitx2* in EOMs.

The T-box gene *Tbx1* is critical for PA muscle development but is not required for trunk myogenesis (Kelly et al., 2004). Unlike *Pitx2*, this transcription factor specifies PA, but not EOM, muscle founder cells (Kelly et al., 2004; Sambasivan et al., 2009). In *Tbx1*-null mouse mutants, first PA myogenesis is impaired, and the posterior arches themselves are absent, highlighting the additional role that *Tbx1* has in ectoderm and endoderm in PA development. Interestingly, a random monolateral ablation of skeletal muscles is often observed in *Tbx1*-null mutants, implicating an all-or-nothing mechanism for the generation of muscle founder cells (Grifone and Kelly, 2007; Kelly et al., 2004). In the progenitors downstream of these founder stem cells, cell fate is assured by cooperation between *Tbx1* and *Myf5*, as PA muscles do not form in these double mutants (Sambasivan et al., 2009). A common feature of PA and EOM founder cells is the epistatic relationship that exists between the MRFs: *Myod* acts genetically downstream of *Tbx1* and *Myf5* in the PA, and downstream of *Myf5* and *Mrf4* in the EOMs.

Tbx1 and *Pitx2* cross-regulate each other and might cooperate to activate the same target genes (Nowotschin et al., 2006), explaining the observation that PA myogenesis is observed occasionally in *Tbx1:Myf5* double mutant mice (Sambasivan et al., 2009). Whether *Pitx2* plays a similar regulatory role to the MRFs in EOM and PA founder cells remains to be determined. As indicated, *Pitx2* and *Tbx1* expression is not restricted to the mesoderm but instead has a much broader expression pattern in cells adjacent to EOM and PA mesoderm, as well as elsewhere. In humans, *PITX2* mutations cause Rieger syndrome, which is associated with ocular and cardiac anomalies and dental hypoplasia (Tumer and Bach-Holm, 2009). Human *TBX1* mutations contribute to the anomalies seen in DiGeorge syndrome, which is characterised by cardiovascular and craniofacial defects together with PA muscle weakness and skeletal muscle hypotonia (Aggarwal and Morrow, 2008). Therefore, these genes could act both cell autonomously, as well as non-cell autonomously, to affect muscle development. That these genes also act cell autonomously to control myogenesis in head mesoderm has been supported by conditional knockout studies in mice (Aggarwal et al., 2010; Dastjerdi et al., 2007; Dong et al., 2006; Grifone and Kelly, 2007; Zacharias et al., 2011). However, additional conditional gene ablation studies are needed to assess cell-cell interactions in regulation of muscle cell fates. Molecular interactions upstream of these two key genes are also yet to be elucidated.

Musculin and Tcf21

Msc (musculin; also known as *MyoR*), which is expressed in head and body muscles, and *Tcf21* (also known as capsulin), are bHLH transcription factors. They are considered to act as repressors although this has not been shown in vivo. They play key roles in the PA, but are not expressed in the EOM founder cells. Mouse embryos with mutations in both these genes fail to develop a subset of first arch-derived jaw muscles (the masseter, pterygoid and temporalis muscles) (Lu et al., 2002). As with *Pitx2* and *Tbx1* mutants, why only a subset of muscles (Table 1) are affected by *Msc* and *Tcf21* deletion remains a mystery. The link between these transcription factors and the MRFs has not been clearly established.

Six family and Eya domain factors

The homeodomain factors *Six1*, *Six4*, *Meox1* and *Meox2*, as well as the Eya domain factors *Eya1* and *Eya2* play critical regulatory roles upstream of *Pax3* in the somitic mesoderm (Table 1). Six, Eya and Pax factors, along with Dachshund family members, act in a network to govern organogenesis in multiple contexts (Relaix and

Buckingham, 1999). However, the role of these factors in the CM, if any, remains to be explored. Critical roles of Six and Eya factors in trunk myogenesis have been demonstrated (Grifone et al., 2007; Grifone et al., 2005). Although *Meox1* and *Meox2* have roles in somite formation and differentiation, intriguingly, subsets of proximal and distal forelimb muscles are missing in *Meox2* mutants, and overall fore- and hindlimb muscle masses are reduced (Mankoo et al., 1999; Mankoo et al., 2003). In fact, Six1 has been implicated in head muscle development in zebrafish (Lin et al., 2009). In spite of this complexity in the gene regulatory network upstream of the MRFs, one could speculate that two or more distinct regulatory cascades at the level of mesoderm specification might have evolved to regulate the MRFs and to orchestrate myogenesis. This scenario might well be true for other lineage programmes in the head versus the trunk.

Taken together, these genetic studies reveal that, although the MRFs have been redeployed to make head muscles, the upstream network that regulates MRF expression seems to have evolved independently in the head.

Temporal changes in myogenic genetic networks

Myogenesis occurs in embryonic, foetal and postnatal phases that are distinguishable by the myogenic cell populations involved and by the gene expression patterns observed during these phases (see Tajbakhsh, 2009). The upstream gene regulatory networks discussed above operate in the embryonic phase of myogenesis. This phase initiates the anlagen, which is built upon during the successive waves of myogenesis. A number of reports, however, suggest that the regulatory programmes that operate in the somite-derived muscle progenitors are different during the embryonic, foetal, juvenile and adult phases of myogenesis (Biressi et al., 2007; Messina et al., 2010; Stockdale, 1992; Tajbakhsh, 2009). A key finding that underscores this view was the discovery that the transcription factor Nfix1 (nuclear factor 1) is a critical regulator of foetal, but not embryonic, myogenesis in mice (Messina et al., 2010). Although nuclear factor protein family members regulate cell fate choices in multiple tissues by acting as repressors or activators, the role of Nfix1 in this context is to repress the embryonic, while promoting the foetal, muscle programme (Messina et al., 2010). How this novel regulator integrates with the early embryonic transcription factors mentioned above, and in different locations, remains to be explored.

Although the genetic hierarchies that act upstream of the MRFs in somitic and cranial mesoderm are distinct, the emergence of a more muscle-lineage restricted, *Pax7*-expressing stem/progenitor population in both the head and the trunk at mid-embryogenesis, always after the establishment of muscle anlage, appears to be common in both cases. This second wave of stem/progenitors in the head and the trunk assures muscle growth until adulthood in the mouse (Kassar-Duchossoy et al., 2005; Relaix et al., 2005). Notably, the ontological molecular signature that characterises embryonic muscle founder cells in different locations is partially retained in the adult stem/progenitors as part of a ‘molecular memory’ (Harel et al., 2009; Sambasivan et al., 2009), hinting at a persistent role for these regulators in the distinct locations.

In summary, a comparison of the distinct regulatory strategies that make muscles in the CM to those that make muscles in the trunk reinforces the view that the CM-derived muscle progenitors are evolutionarily distinct from the somitic muscle progenitor pool. An assessment of the key molecular players that are divergent between head and trunk myogenic programmes in an evolutionary context is pertinent in this context.

Evolutionary origins of cranial muscle

The distinct organisation of CM and the divergent gene networks employed by the muscle progenitors in CM to make head muscles provide clues to the evolutionary origins of craniofacial muscles. This section analyses comparative morphology, as well as the expression of key genes, discussed above, in select model organisms in order to understand vertebrate head muscle evolution.

It is likely that the ancestor to the vertebrates looked more like the extant amphioxus than the more closely related tunicate (a urochordate), as tunicates do not have somites (Yu et al., 2007). In the amphioxus, the unsegmented CM is not present, raising the possibility that some of the anterior somites (of visceral mesoderm origin) in the amphioxus-like ancestor lost their segmental configuration, resulting in the non-segmented head mesoderm of vertebrates (Holland et al., 2008). Although overt branchial arch muscles are absent in the adult amphioxus, a distinction between the branchiomeric (see Glossary, Box 1) and somitomeric muscles might, nevertheless, exist (Ruppert, 1997). This is because the so-called pterygial muscles, which develop more ventrally in the amphioxus pharynx (Fig. 4A), are innervated by the peripheral nerves that resemble the branchiomeric nerves in vertebrates (Fritzsch and Northcutt, 1993). This implies that, even before the origin of the vertebrate-specific CM, the PA mesoderm-like cell lineages might have been present in the vertebrate ancestor and that during evolution they differentiated into muscles that could be innervated by specific sets of peripheral nerves distinct from those innervating the myotomes in the trunk. Furthermore, *AmphiTbx1/10* (a single gene that is the forerunner of the vertebrate *Tbx1* and *Tbx10* genes) is expressed in the PA mesoderm of amphioxus (Mahadevan et al., 2004). Taken together, these findings suggest that the cell population and the genetic machinery that distinguish head muscles from those of somitic mesoderm origin might have existed, in part, long before the evolution of the vertebrate head. Concordantly, in the basal jawless vertebrate lamprey, *Tbx1/10* is expressed in the mesodermal core of the PAs (Sauka-Spengler et al., 2002; Tiecke et al., 2007), as well as in the labial and velar (mandibular arch-derived muscles) muscle progenitors. Furthermore, although *Caenorhabditis elegans tbx-2* is not closely related to murine *Tbx1* (Larroux et al., 2008), it was shown that *tbx-2* is required for the development of the *C. elegans* pharyngeal muscles, but only those located anteriorly (Smith and Mango, 2007). It is tempting, therefore, to speculate that the ancestral *Tbx1* gene might have acquired a myogenic task in the CM early in evolution.

Whole-genome comparative data have shown that, unlike the Pax genes, for which a *PaxB*-like gene founded the Pax class before the separation of sponges and eumetazoans (all major animal groups), the T-box transcription factors *T* (*Brachyury*), *Tbx 4*/*5* (precursor of the paralogous group *Tbx4* and *Tbx5*) and *Tbx1/15/20* were present in the last common ancestor of metazoans (Larroux et al., 2008). With respect to the MRFs, a single ancestor was proposed to be conserved from *Drosophila* to jellyfish to tunicates (Atchley et al., 1994), undergoing two rounds of duplication events to yield the current MRFs in vertebrates. As for *Pitx2*, the *AmphiPitx* gene is not expressed in the expected region in the developing larval head (Yasui et al., 2000), but the lamprey *Pitx* (orthologue of *Pitx1* or *Pitx2*) is expressed in the pre-mandibular mesoderm that apparently gives rise to EOMs (Boorman and Shimeld, 2002; Kusakabe and Kuratani, 2007). However, the development of the lamprey EOMs is not yet well understood, therefore, further investigation of lamprey craniofacial development could lead to a better understanding of EOM ontology in vertebrates.

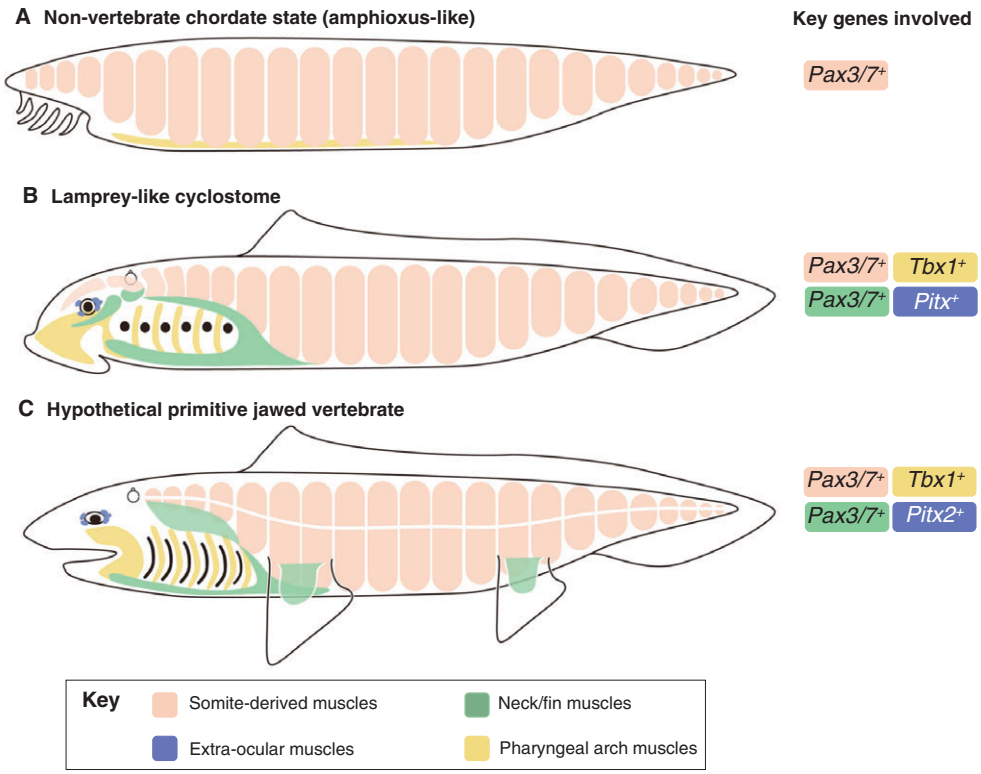

Fig. 4. Hypothetical evolutionary scheme of different muscle groups in chordates. Schematic of muscle and gene evolution in (**A**) an amphioxus-like chordate ancestor; (**B**) a lamprey-like cyclostome; and (**C**) a hypothetical primitive jawed vertebrate. (A) In the amphioxus-like ancestor, *Pax3*/*Pax7*-positive myotomes (pink) are present along the anteroposterior (AP) axis and a putative precursor of branchial muscle (yellow) is present in the pharyngeal floor. (B) In the lamprey-like cyclostome, extra-ocular muscles (EOMs; purple) and pharyngeal arch (PA) muscles (yellow) develop from head mesoderm that express *Pitx* and *Tbx1/10*, respectively. Poorly differentiated cucullaris and hypobrancial muscles (green) derived from migratory founder cells are also present. Note that anterior myotomes in the cyclostome originally develop from postotic somites and secondarily migrate rostrally during development. (C) In a hypothetical jawed vertebrate, migratory founders have differentiated, establishing the neck, as well as paired fin muscles. Also note that myotomes are dorsoventrally compartmentalised into epaxial and hypaxial muscles in jawed vertebrates. The horizontal white line represents the epaxial/hypaxial boundary separating the dorsal/ventral portions of the myotomes. Modified with permission from Kusakabe and Kuratani (Kusakabe and Kuratani, 2005). Hypothetical or known expression patterns of the critical transcription factor orthologues (of Pax3/7, Tbx1 and Pitx2) in the progenitors of distinct muscle groups are also indicated.

As mentioned previously, the *Pax3* and *Pax7* genes have been associated with the myogenic development of the vertebrate somitic mesoderm (Buckingham and Relaix, 2007; Sambasivan and Tajbakhsh, 2007). In the early stages of vertebrate evolution, *Pax3* and *Pax7* were involved in dorsal specification of the neural tube and the acquisition of neural crest, as well as in the compartmentalisation of myotomes. A Pax3/7-positive layer of cells in some bony fish embryos and even in lamprey embryos appears to be the homologue of the epithelial cells of the dermomyotome present in amniotes (Kusakabe and Kuratani, 2007) and in zebrafish (Hollway et al., 2007; Stellabotte et al., 2007). In amphioxus too, *AmphiPax3/7* (ancestor of *Pax3* and *Pax7*) could play a role in myogenic development, as it is expressed in the anterior and posterior somites (Holland et al., 1999). If the hypothesis that the anterior somites are the ancestors of vertebrate CM is true (Holland et al., 2008), the evolution of the CM would involve not only the loss of segmentation but also a radical shift in its molecular regulation, i.e. loss of the *Pax3* myogenic pathway for *Tbx1* and/or *Pitx2* regulatory cascades. Alternatively, as suggested above, the CM might have originated as an evolutionarily novel tissue from an entirely different cell population, such as the more ventral cell lineage that expresses *AmphiTbx1*.

It is interesting to note that in the vertebrate gastrula, the CM population arises from the primitive streak prior to the trunk mesoderm, yet myogenic induction in the head is delayed compared with that observed in the trunk. One could speculate that this lag in differentiation might correlate with the later phylogenetic appearance of head muscles. Molecular data from across the phylogenetic spectrum support the notion that myogenic potential was acquired by distinct embryonic cell populations, and that, in the anterior mesoderm, this population evolved independent strategies with respect to the posterior mesoderm to accomplish MRF induction and myogenesis. Therefore, whereas the tongue and some neck muscles might have evolved secondarily as an extension of the trunk programme, the CM-derived head muscles are generally viewed to have evolved independently.

Neck muscles: a transition zone

The neck region provides interesting correlative information because it is a transition zone between the trunk and head. Typically, as seen in the region anterior to the otic placode in the head of many vertebrate embryos, only the ventral portion of the CM is divided segmentally into PAs whereas the dorsal paraxial portion is unsegmented. By contrast, the neck region is characterised by the presence of both somites and pharyngeal

mesoderm (Fig. 3). As a result, two different types of segmentation appear in this transition zone, as seen in the large part of the body axis in the amphioxus. Furthermore, the peripheral morphology of the cranial nerves show the highest level of complexity in this region (Kuratani, 1997).

The neck muscles are known collectively as the hypobranchial and cucullaris muscle groups (see Glossary, Box 1). Some of these muscles arise as migratory founder cells from an anterior group of occipital somites and some from cervical somites (Birchmeier and Brohmann, 2000), although the origin of the cucullaris group has been contested (Huang et al., 2001; Kusakabe and Kuratani, 2005; Noden and Francis-West, 2006; Theis et al., 2010). Of note, ectomesenchyme derived from cranial neural crest cells contributes to connective tissues and influences the morphological patterning of muscles in the domain spanning the posterior cranium and the shoulder girdle (Huang et al., 2006; Matsuoka et al., 2005; McKenzie, 1962; Noden, 1983a; Rinon et al., 2007), such as the trapezius, the sternocleidomastoid, the infrahydoid muscle group, the floor of the oral cavity, the tongue muscles and the diaphragm in mammals. Most of these muscles also share a curious trait in that they are not associated with the body axis or the lateral body wall, as typical skeletal muscles are, but instead are associated with visceral components of the body, such as the digestive tract and the pharynx. Notably, the hypobranchial muscles do not form as a continuous growth of muscle plates, but as migrating myoblasts along the non-segmental pathway. Furthermore, they are not found in the lateral body wall as normally expected for trunk muscles. In this regard, lamprey hypobranchial muscle also shares some traits with the neck muscle in gnathostomes in that it arises from somites and attaches to branchial arch skeletons. The contribution of the cephalic crest cells to this muscle, however, is not known in this animal.

Genetic data suggest that some of the neck muscles are more similar to head muscles than to trunk muscles, as *Pax3: Myf5 (Mrf4)*-null mouse mutants that lack somite-derived musculature retain trapezius and sternocleidomastoid (cucullaris group) muscles (Tajbakhsh et al., 1997; Theis et al., 2010), whereas *Tbx1*$^{-/-}$ mouse mutants that lack the first and second PA-derived muscles also lack the trapezius and sternocleidomastoid muscles (Kelly et al., 2004; Theis et al., 2010). Although earlier reports showed a somitic origin for cucullaris muscles (Huang et al., 2001; Noden and Francis-West, 2006), a recent study has reported that these muscles have a non-somitic origin (Theis et al., 2010) and concurred with a head muscle programme for cucullaris muscle development. A closer examination of the embryological origins of these muscles is warranted given that *Pax3*, which affects somite-derived muscles, does not appear to regulate some muscles of the neck, indicating that multiple origins might, in fact, define this complex transition zone.

Ancestral vertebrates do not seem to have possessed limb or neck muscles, thus cyclostomes (lampreys and hagfish, see Glossary, Box 1), which also lack typical neck and hypobranchial muscles, are often used as model organisms in which to investigate the evolutionary origins of this musculature. In the lamprey, a muscle called the ‘hypobranchial’ muscle does exist and it is innervated by the hypoglossal nerve. However, this muscle does not invade into the oral floor as it does in gnathostomes (Kuratani et al., 1999). It might represent, therefore, an evolutionary precursor for the neck muscles. Modern cyclostomes do not possess paired fins or shoulder girdles, either. As there is no information on the later contributions of cranial neural crest cells in the lamprey, it is more challenging to define a ‘neck’ domain in this animal (Kuratani, 2008a). Thus, evolution of the gnathostomes, after the split from cyclostomes, would have been initiated by the acquisition of the shoulder girdle. This, in turn, might have been associated with and/or based upon the establishment of cucullaris and hypobranchial muscles (as well as tongue, infrahyoid muscle groups and possibly the diaphragm) in the neck together with the posterior expansion of the cranial neural crest cells that function in the patterning of these muscles. This raises the question: which arose first in evolution – neck or pectoral fin muscles?

Pax3 and *Lbx1* (ladybird homeobox 1) are expressed in migratory founder cells of the hypobranchial muscles and therefore might be necessary for their ontogeny in vertebrates (Neyt et al., 2000), although these muscles are present in *Lbx1* mouse mutants (Gross et al., 2000). The presence of a primitive form of hypobranchial muscle in the lamprey suggests that the neck muscle began to differentiate before the acquisition of the paired fin muscle, even in the evolutionary state in which an epaxial/hypaxial (dorsal and ventral to the horizontal septum, respectively) distinction had not been obtained clearly in the myotomes (Kusakabe and Kuratani, 2005; Kusakabe and Kuratani, 2007; Kusakabe et al., 2011). Therefore, further molecular and comparative studies of the lamprey could provide additional insights into the developmental history of neck and fin muscle evolution in vertebrates.

Cranial mesoderm duality: skeletal and cardiac muscle fates

A remarkable feature of the branchiomeric subset of head muscles is its relationship with cardiomyogenesis. The cranial lateral mesoderm supplies founder cells to the jaw and facial muscles, as well as to the right ventricle and outflow tract (OFT, see Glossary, Box 1) of the heart (Fig. 2). Importantly, a subset of cranial neural crest cells also contributes to heart development (Hutson and Kirby, 2007; Hutson et al., 2009). Hence, it is not clear whether cells in the cranial lateral mesoderm are bipotent, if there are distinct committed cardiomyogenic and myogenic founder cells in this compartment, or to what extent cranial muscle and heart programmes are molecularly related.

Lineage-tracing analysis and genetic studies have revealed that molecular overlaps exist between heart and head muscle development. Both Tbx1 and Pitx2, the hierarchical transcription factors in the gene regulatory networks that orchestrate myogenesis in the PA, are known to control the proliferation of cardiac founder cells, and they are required for normal OFT development (Grifone and Kelly, 2007; Tzahor, 2009). *Tcf21* and *Msc*, which are key players in jaw myogenesis (Lu et al., 2002), are also expressed in myocardial founder cells (Robb et al., 1998; von Scheven et al., 2006b). Islet1 (Isl1) is a LIM homeodomain transcription factor and Mef2c is a MADS box transcription factor, and lineage analysis using *Isl1*^{Cre} and *Tg:Mef2c-Cre* reporter mice has revealed that PA-derived muscles have a history of expression of these critical regulators of the heart programme (Abu-Issa and Kirby, 2007; Cai et al., 2003; Dong et al., 2006; Kwon et al., 2009; Mann et al., 2009; Nathan et al., 2008; Shih et al., 2007; Verzi et al., 2005). Finally, retrospective clonal lineage analysis in mouse embryos has revealed the existence of bipotent ancestral cells, either in the cranial lateral mesoderm or even earlier during development, that give rise to both arch-derived skeletal muscles and anterior heart field-derived cardiomyocytes (Lescroart et al., 2010). Interestingly, this report also shows the lineage relationships between first PA-derived muscles and the right ventricle, and the second PA-derived muscles and the OFT, indicating diversity at the

cellular level between first- and second-arch muscle progenitors. In fact, the first and second pharyngeal arches are likely to have distinct molecular programmes, as supported by the phenotypes of *Tbx1*-null mice (in which the first arch is present, whereas other arches are absent) (Jerome and Papaioannou, 2001) and *Pitx2* mutant mice (in which only the first arch is hypoplastic) (Gage et al., 1999; Shih et al., 2007).

Nkx2.5, another key homeodomain-containing transcription factor in the cardiomyogenic programme (Harvey et al., 2002), is expressed early in arch-derived pterygoid muscle in vertebrates (Kasahara et al., 1998). Furthermore, targeting of the *Nkx2.5* locus with Cre recombinase has allowed its expression in these muscles to be traced in mice (Kasahara et al., 1998; Stanley et al., 2002). In addition, in *C. elegans*, in which cardiac tissue is not present and the pharynx exhibits electrical activity, the *Nkx2.5* homologue *ceh-22* is expressed during the differentiation of pharyngeal muscle (Okkema and Fire, 1994). In the tunicate *Ciona intestinalis*, cells derived from *NK4* (*Nkx2.5* homologue)-expressing blastomeres generate the heart, as well as the muscles of the atrial siphon (the cloacal siphon in the body-wall) (Hirano and Nishida, 1997; Stolfi et al., 2010). Interestingly, *Islet* (the *Ciona* homologue of *Isl1*) is not expressed in heart progenitors but it marks the atrial siphon muscle lineage (Stolfi et al., 2010). This report also shows that some cells of the longitudinal muscles, which arise from atrial siphon muscles upon metamorphosis, also express *Tbx1/10* (the gene ancestral to *Tbx1* and *Tbx10*).

Notably, the promiscuous expression of molecular markers in cardiomyogenic and skeletal myogenic founder cells also extends to their differentiated progeny. For example, human and rabbit masticatory muscles retain cardiac α-myosin heavy chain expression (Bredman et al., 1991), as do EOMs in rabbits (Rushbrook et al., 1994). However, cardiac α-myosin heavy chain is not expressed in the jaw muscles of the rat (Kawai et al., 2009). Other evidence that cranial paraxial mesoderm retains a kinship with cranial lateral mesoderm comes from manipulations in the chick, in which ectopic bone morphogenetic protein 4, Bmp4, induces cardiac markers and suppresses skeletal muscle markers in CM (Tzahor et al., 2003). Strikingly, this is also the case with jaw, but not limb, muscle satellite cells, in which cardiac markers (e.g. *Isl1* and *Tbx20*) are activated upon Bmp4 induction (Harel et al., 2009).

Thus, the hierarchical transcription factors that regulate muscle development in PAs are expressed in cardiac progenitors and vice versa. The overlap of hierarchical gene expression in founder cells of the cardiac and head muscle lineages raises the following question: what mechanisms define these alternate myogenic fates? Canonical Wnt signalling, which promotes somitic muscle development, is inhibitory to both cardiac and branchiomeric muscle differentiation (Cossu and Borello, 1999; Münsterberg et al., 1995; Nathan et al., 2008; Stern and Hauschka, 1995; Tajbakhsh et al., 1998; Tzahor et al., 2003; Tzahor and Lassar, 2001). By contrast, the BMP pathway, a negative influence on trunk muscle development, also acts negatively on head myogenesis (Pourquié et al., 1996; Tirosh-Finkel et al., 2006; Tzahor et al., 2003; von Scheven et al., 2006a) but it promotes cardiomyogenic differentiation (Dyer and Kirby, 2009; Tirosh-Finkel et al., 2006; von Scheven et al., 2006a). Furthermore, sonic hedgehog (Shh), which promotes trunk muscle development (Borycki et al., 1999), has been shown to act as a negative cue for branchiomeric myogenesis (Tzahor et al., 2003; von Scheven et al., 2006a). By contrast, Shh has been implicated in regulating *Tbx1* expression in the PAs (Garg et al., 2001). The role of Shh in cardiomyogenic differentiation is not clear; however, it is critically required for progenitor cell proliferation and heart tube morphogenesis (Dyer and Kirby, 2009; Washington Smoak et al., 2005). Another influential signalling molecule, fibroblast growth factor 8 (Fgf8), inhibits skeletal myogenic differentiation (von Scheven et al., 2006a), but promotes the cardiogenic programme (Dyer and Kirby, 2009). However, BMP-mediated inhibition of Fgf signalling has been shown to promote cardiomyocyte differentiation (Tirosh-Finkel et al., 2010).

In essence, the genes that are critical for head myogenesis, such as *Tbx1*, *Pitx2*, *Tcf21* and *Msc*, and that distinguish it from the trunk muscle programme, are shared with the cardiogenesis developmental programme. This feature further underscores the theme highlighted in this review: that the head muscles are evolutionarily distinct from trunk muscles, and that the head myogenic gene network probably arose independently of the trunk muscle regulatory programme.

Conclusions

The convergence of developmental and evolutionary biology with comparative genomic data has filled in some of the gaps in our knowledge about the CM and its derivatives. A major challenge, however, is to determine when and how the CM appeared in the ancestor of vertebrates as a trait specific to vertebrates, how it became regionalised and when this coincided with the emergence of founder stem cells for skeletal and cardiac muscle, the skull bones and endothelium. An intriguing thought, from an evo-devo perspective, is whether CM patterning and differentiation mechanisms could be viewed as a modification of the myotome developmental mechanism, which is putatively ancestral to the CM. In this regard, distinguishing between the paraxial and lateral components of the CM is a fundamental issue. At present, morphological criteria are not sufficient to distinguish the cranial prechordal, paraxial and lateral components of the CM, and gene expression analyses have not yet identified specific markers for each. When this becomes possible, genetic lineage studies using an inducible Cre recombinase with reporter mice might clarify some of these issues. Another question is whether or not there is indeed a ‘paraxial’ and ‘lateral’ mesoderm, as is distinguished in the trunk, and, if so, to what extent these tissues are limited in fate potential. This remains a matter of debate. The overlap in expression of cardiac and skeletal myogenic transcription factors in each suggests that these fates are not as distinct as originally thought. Repressive signatures might also delay cell fate acquisition and allow proliferation until specific cell lineages arrive at their proper destination. Identifying novel regulators and elucidating the crosstalk between the factors that have been shown to be critical, including Tbx1, Pitx2 and the MRFs, will also be major immediate goals.

In the context of the evolution of the CM, some major shifts in thinking and classification have been proposed in the last five years, based on studies of tunicates, amphioxus and lampreys, all of which have provided informative clues as to how the CM evolved. For a true comparative analysis, it is important to consider the acquisition, as well as the loss, of traits; however, this adds to the complexity of these studies. Nevertheless, neural crest, cranial nerves and the evolution of the neck muscles provide landmarks for understanding how CM organisation and cell fate occur. Gene expression profiles of critical transcription factors in the ancestral model organisms have also been insightful in determining the developmental state of anterior mesoderm. Transplantation and cell lineage approaches in avian and mouse models have been arguably

the most informative in unravelling the complexities of CM fates. Applying these types of experimental approaches to cyclostomes and gnathostomes should provide the missing pieces of the puzzle.

Acknowledgements
We are grateful to Robert Kelly for critical comments. S.T. is funded by the Institut Pasteur, Centre National de la Recherche Scientifique, Association Française contre le Myopathies, Agence Nationale de la Recherche, EU Framework 7 projects EuroSysStem and Optistem, and the Fondation pour la Recherche Medicale. S.K. acknowledges Kinya Yasui and Rie Kusakabe for valuable discussions.

References

Abu-Issa, R. and Kirby, M. L. (2007). Heart field: from mesoderm to heart tube. *Annu. Rev. Cell Dev. Biol.* **23**, 45-68.
Aggarwal, V. S. and Morrow, B. E. (2008). Genetic modifiers of the physical malformations in velo-cardio-facial syndrome/DiGeorge syndrome. *Dev. Disabil. Res. Rev.* **14**, 19-25.
Aggarwal, V. S., Carpenter, C., Freyer, L., Liao, J., Petti, M. and Morrow, B. E. (2010). Mesodermal Tbx1 is required for patterning the proximal mandible in mice. *Dev. Biol.* **344**, 669-681.
Atchley, W. R., Fitch, W. M. and Bronner-Fraser, M. (1994). Molecular evolution of the MyoD family of transcription factors. *Proc. Natl. Acad. Sci. USA* **91**, 11522-11526.
Balfour, F. M. (1878). The development of the elasmo-branch fishes. *J. Anat. Physiol.* **12**, 177-216.
Beaster-Jones, L., Horton, A. C., Gibson-Brown, J. J., Holland, N. D. and Holland, L. Z. (2006). The amphioxus T-box gene, AmphiTbx15/18/22, illuminates the origins of chordate segmentation. *Evol. Dev.* **8**, 119-129.
Bergstrom, D. A. and Tapscott, S. J. (2001). Molecular distinction between specification and differentiation in the myogenic basic helix-loop-helix transcription factor family. *Mol. Cell. Biol.* **21**, 2404-2412.
Birchmeier, C. and Brohmann, H. (2000). Genes that control the development of migrating muscle precursor cells. *Curr. Opin. Cell Biol.* **12**, 725-730.
Biressi, S., Molinaro, M. and Cossu, G. (2007). Cellular heterogeneity during vertebrate skeletal muscle development. *Dev. Biol.* **308**, 281-293.
Bober, E., Franz, T., Arnold, H. H., Gruss, P. and Tremblay, P. (1994). Pax-3 is required for the development of limb muscles: a possible role for the migration of dermomyotomal muscle progenitor cells. *Development* **120**, 603-612.
Boorman, C. J. and Shimeld, S. M. (2002). Cloning and expression of a Pitx homeobox gene from the lamprey, a jawless vertebrate. *Dev. Genes Evol.* **212**, 349-353.
Borycki, A. G., Brunk, B., Tajbakhsh, S., Buckingham, M., Chiang, C. and Emerson, C. P., Jr (1999). Sonic hedgehog controls epaxial muscle determination through Myf5 activation. *Development* **126**, 4053-4063.
Bothe, I. and Dietrich, S. (2006). The molecular setup of the avian head mesoderm and its implication for craniofacial myogenesis. *Dev. Dyn.* **235**, 2845-2860.
Bredman, J. J., Wessels, A., Weijs, W. A., Korfage, J. A., Soffers, C. A. and Moorman, A. F. (1991). Demonstration of 'cardiac-specific' myosin heavy chain in masticatory muscles of human and rabbit. *Histochem. J.* **23**, 160-170.
Buckingham, M. and Relaix, F. (2007). The role of Pax genes in the development of tissues and organs: Pax3 and Pax7 regulate muscle progenitor cell functions. *Annu. Rev. Cell Dev. Biol.* **23**, 645-673.
Cai, C. L., Liang, X., Shi, Y., Chu, P. H., Pfaff, S. L., Chen, J. and Evans, S. (2003). Isl1 identifies a cardiac progenitor population that proliferates prior to differentiation and contributes a majority of cells to the heart. *Dev. Cell* **5**, 877-889.
Chen, Y. H. and Tsai, H. J. (2002). Treatment with Myf5-morpholino results in somite patterning and brain formation defects in zebrafish. *Differentiation* **70**, 447-456.
Cheng, G., Merriam, A. P., Gong, B., Leahy, P., Khanna, S. and Porter, J. D. (2004). Conserved and muscle-group-specific gene expression patterns shape postnatal development of the novel extraocular muscle phenotype. *Physiol. Genomics* **18**, 184-195.
Cordes, S. P. (2001). Molecular genetics of cranial nerve development in mouse. *Nat. Rev. Neurosci.* **2**, 611-623.
Cossu, G. and Borello, U. (1999). Wnt signaling and the activation of myogenesis in mammals. *EMBO J.* **18**, 6867-6872.
Couly, G. F., Coltey, P. M. and Douarin, N. M. L. (1992). The developmental fate of the cephalic mesoderm in quail-chick chimeras. *Development* **114**, 1-15.
Couly, G. F., Coltey, P. M. and Le Douarin, N. M. (1993). The triple origin of skull in higher vertebrates: a study in quail-chick chimeras. *Development* **117**, 409-429.
Creuzet, S., Couly, G., Vincent, C. and Le Douarin, N. M. (2002). Negative effect of Hox gene expression on the development of the neural crest-derived facial skeleton. *Development* **129**, 4301-4313.
Dastjerdi, A., Robson, L., Walker, R., Hadley, J., Zhang, Z., Rodriguez-Niedenfuhr, M., Ataliotis, P., Baldini, A., Scambler, P. and Francis-West, P. (2007). Tbx1 regulation of myogenic differentiation in the limb and cranial mesoderm. *Dev. Dyn.* **236**, 353-363.
Dong, F., Sun, X., Liu, W., Ai, D., Klysik, E., Lu, M. F., Hadley, J., Antoni, L., Chen, L., Baldini, A. et al. (2006). Pitx2 promotes development of splanchnic mesoderm-derived branchiomeric muscle. *Development* **133**, 4891-4899.
Dubrulle, J. and Pourquie, O. (2002). From head to tail: links between the segmentation clock and antero-posterior patterning of the embryo. *Curr. Opin. Genet. Dev.* **12**, 519-523.
Dyer, L. A. and Kirby, M. L. (2009). The role of secondary heart field in cardiac development. *Dev. Biol.* **336**, 137-144.
Ericsson, R., Cerny, R., Falck, P. and Olsson, L. (2004). Role of cranial neural crest cells in visceral arch muscle positioning and morphogenesis in the Mexican axolotl, Ambystoma mexicanum. *Dev. Dyn.* **231**, 237-247.
Evans, D. J. and Noden, D. M. (2006). Spatial relations between avian craniofacial neural crest and paraxial mesoderm cells. *Dev. Dyn.* **235**, 1310-1325.
Fischer, M. D., Gorospe, J. R., Felder, E., Bogdanovich, S., Pedrosa-Domellof, F., Ahima, R. S., Rubinstein, N. A., Hoffman, E. P. and Khurana, T. S. (2002). Expression profiling reveals metabolic and structural components of extraocular muscles. *Physiol. Genomics* **9**, 71-84.
Franz, T., Kothary, R., Surani, M. A., Halata, Z. and Grim, M. (1993). The Splotch mutation interferes with muscle development in the limbs. *Anat. Embryol.* **187**, 153-160.
Fritzsch, B. and Northcutt, R. G. (1993). Cranial and spinal nerve organization in amphioxus and lampreys: evidence for an ancestral craniate pattern. *Acta Anat. (Basel)* **148**, 96-109.
Gage, P. J., Suh, H. and Camper, S. A. (1999). Dosage requirement of Pitx2 for development of multiple organs. *Development* **126**, 4643-4651.
Gage, P. J., Rhoades, W., Prucka, S. K. and Hjalt, T. (2005). Fate maps of neural crest and mesoderm in the mammalian eye. *Invest. Ophthalmol. Vis. Sci.* **46**, 4200-4208.
Gans, C. and Northcutt, R. G. (1983). Neural crest and the origin of vertebrates: a new head. *Science* **220**, 268-273.
Garg, V., Yamagishi, C., Hu, T., Kathiriya, I. S., Yamagishi, H. and Srivastava, D. (2001). Tbx1, a DiGeorge syndrome candidate gene, is regulated by sonic hedgehog during pharyngeal arch development. *Dev. Biol.* **235**, 62-73.
Goodrich, L. V. (1930). *Studies on the Structure and Development of Vertebrates*. London: McMillan.
Goulding, M., Lumsden, A. and Paquette, A. J. (1994). Regulation of Pax-3 expression in the dermomyotome and its role in muscle development. *Development* **120**, 957-971.
Grammatopoulos, G. A., Bell, E., Toole, L., Lumsden, A. and Tucker, A. S. (2000). Homeotic transformation of branchial arch identity after Hoxa2 overexpression. *Development* **127**, 5355-5365.
Grifone, R. and Kelly, R. G. (2007). Heartening news for head muscle development. *Trends Genet.* **23**, 365-369.
Grifone, R., Demignon, J., Houbron, C., Souil, E., Niro, C., Seller, M. J., Hamard, G. and Maire, P. (2005). Six1 and Six4 homeoproteins are required for Pax3 and Mrf expression during myogenesis in the mouse embryo. *Development* **132**, 2235-2249.
Grifone, R., Demignon, J., Giordani, J., Niro, C., Souil, E., Bertin, F., Laclef, C., Xu, P. X. and Maire, P. (2007). Eya1 and Eya2 proteins are required for hypaxial somitic myogenesis in the mouse embryo. *Dev. Biol.* **302**, 602-616.
Gross, M. K., Moran-Rivard, L., Velasquez, T., Nakatsu, M. N., Jagla, K. and Goulding, M. (2000). Lbx1 is required for muscle precursor migration along a lateral pathway into the limb. *Development* **127**, 413-424.
Hacker, A. and Guthrie, S. (1998). A distinct developmental programme for the cranial paraxial mesoderm in the chick embryo. *Development* **125**, 3461-3472.
Hamburger V. and Hamilton H. L. (1992). A series of normal stages in the development of the chick embryo. 1951. *Dev. Dyn.* **195**, 231-272.
Harel, I., Nathan, E., Tirosh-Finkel, L., Zigdon, H., Guimaraes-Camboa, N., Evans, S. M. and Tzahor, E. (2009). Distinct origins and genetic programs of head muscle satellite cells. *Dev. Cell* **16**, 822-832.
Harvey, R. P., Lai, D., Elliott, D., Biben, C., Solloway, M., Prall, O., Stennard, F., Schindeler, A., Groves, N., Lavulo, L. et al. (2002). Homeodomain factor Nkx2-5 in heart development and disease. *Cold Spring Harb. Symp. Quant. Biol.* **67**, 107-114.
Hasty, P., Bradley, A., Morris, J. H., Edmondson, D. G., Venuti, J. M., Olson, E. N. and Klein, W. H. (1993). Muscle deficiency and neonatal death in mice with a targeted mutation in the myogenin gene. *Nature* **364**, 501-506.
Hinits, Y., Osborn, D. P. and Hughes, S. M. (2009). Differential requirements for myogenic regulatory factors distinguish medial and lateral somitic, cranial and fin muscle fibre populations. *Development* **136**, 403-414.
Hirano, T. and Nishida, H. (1997). Developmental fates of larval tissues after metamorphosis in ascidian Halocynthia roretzi. I. Origin of mesodermal tissues of the juvenile. *Dev. Biol.* **192**, 199-210.
Holland, L. Z., Schubert, M., Kozmik, Z. and Holland, N. D. (1999). AmphiPax3/7, an amphioxus paired box gene: insights into chordate myogenesis, neurogenesis, and the possible evolutionary precursor of definitive vertebrate neural crest. *Evol. Dev.* **1**, 153-165.

Holland, L. Z., Holland, N. D. and Gilland, E. (2008). Amphioxus and the evolution of head segmentation. *Integr. Comp. Biol.* **48**, 630-646.
Holland, P. W. (2000). Embryonic development of heads, skeletons and amphioxus: Edwin S. Goodrich revisited. *Int. J. Dev. Biol.* **44**, 29-34.
Hollway, G. E., Bryson-Richardson, R. J., Berger, S., Cole, N. J., Hall, T. E. and Currie, P. D. (2007). Whole-somite rotation generates muscle progenitor cell compartments in the developing zebrafish embryo. *Dev. Cell* **12**, 207-219.
Hu, P., Geles, K. G., Paik, J. H., DePinho, R. A. and Tjian, R. (2008). Codependent activators direct myoblast-specific MyoD transcription. *Dev. Cell* **15**, 534-546.
Huang, R., Lang, E. R., Otto, W. R., Christ, B. and Patel, K. (2001). Molecular and cellular analysis of embryonic avian tongue development. *Anat. Embryol. (Berl.)* **204**, 179-187.
Huang, R., Christ, B. and Patel, K. (2006). Regulation of scapula development. *Anat. Embryol. (Berl.)* **211 Suppl. 1**, 65-71.
Hutson, M. R. and Kirby, M. L. (2007). Model systems for the study of heart development and disease. Cardiac neural crest and conotruncal malformations. *Semin. Cell Dev. Biol.* **18**, 101-110.
Hutson, M. R., Sackey, F. N., Lunney, K. and Kirby, M. L. (2009). Blocking hedgehog signaling after ablation of the dorsal neural tube allows regeneration of the cardiac neural crest and rescue of outflow tract septation. *Dev. Biol.* **335**, 367-373.
Jacob, M., Jacob, H. J., Wachtler, F. and Christ, B. (1984). Ontogeny of avian extrinsic ocular muscles. I. A light- and electron-microscopic study. *Cell Tissue Res.* **237**, 549-557.
Jacobson, A. G. (1988). Somitomeres: mesodermal segments of vertebrate embryos. *Development* **104**, 209-220.
Jacobson, A. G. (1993). Somitomeres: mesodermal segments of the head and trunk. In *The Skull*, vol. 1 (ed. J. Hanken, and B. K. Hall), pp. 42-76. Chicago: Chicago Press.
Jarvik, E. (1980). *Basic Structure and Evolution of Vertebrates*. New York: Academic Press.
Jerome, L. A. and Papaioannou, V. E. (2001). DiGeorge syndrome phenotype in mice mutant for the T-box gene, Tbx1. *Nat. Genet.* **27**, 286-291.
Jouve, C., Iimura, T. and Pourquie, O. (2002). Onset of the segmentation clock in the chick embryo: evidence for oscillations in the somite precursors in the primitive streak. *Development* **129**, 1107-1117.
Kasahara, H., Bartunkova, S., Schinke, M., Tanaka, M. and Izumo, S. (1998). Cardiac and extracardiac expression of Csx/Nkx2.5 homeodomain protein. *Circ. Res.* **82**, 936-946.
Kassar-Duchossoy, L., Gayraud-Morel, B., Gomès, D., Rocancourt, D., Buckingham, M., Shinin, V. and Tajbakhsh, S. (2004). Mrf4 determines skeletal muscle identity in Myf5:Myod double-mutant mice. *Nature* **431**, 466-471.
Kassar-Duchossoy, L., Giacone, E., Gayraud-Morel, B., Jory, A., Gomes, D. and Tajbakhsh, S. (2005). Pax3/Pax7 mark a novel population of primitive myogenic cells during development. *Genes Dev.* **19**, 1426-1431.
Kawai, N., Sano, R., Korfage, J. A., Nakamura, S., Tanaka, E., van Wessel, T., Langenbach, G. E. and Tanne, K. (2009). Functional characteristics of the rat jaw muscles: daily muscle activity and fiber type composition. *J. Anat.* **215**, 656-662.
Kelly, R. G. (2010). Core issues in craniofacial myogenesis. *Exp. Cell Res.* **316**, 3034-3041.
Kelly, R. G., Brown, N. A. and Buckingham, M. E. (2001). The arterial pole of the mouse heart forms from Fgf10-expressing cells in pharyngeal mesoderm. *Dev. Cell* **1**, 435-440.
Kelly, R. G., Jerome-Majewska, L. A. and Papaioannou, V. E. (2004). The del22q11.2 candidate gene Tbx1 regulates branchiomeric myogenesis. *Hum. Mol. Genet.* **13**, 2829-2840.
Kitamura, K., Miura, H., Miyagawa-Tomita, S., Yanazawa, M., Katoh-Fukui, Y., Suzuki, R., Ohuchi, H., Suehiro, A., Motegi, Y., Nakahara, Y. et al. (1999). Mouse Pitx2 deficiency leads to anomalies of the ventral body wall, heart, extra- and periocular mesoderm and right pulmonary isomerism. *Development* **126**, 5749-5758.
Knapp, J. R., Davie, J. K., Myer, A., Meadows, E., Olson, E. N. and Klein, W. H. (2006). Loss of myogenin in postnatal life leads to normal skeletal muscle but reduced body size. *Development* **133**, 601-610.
Köntges, G. and Lumsden, A. (1996). Rhombencephalic neural crest segmentation is preserved throughout craniofacial ontogeny. *Development* **122**, 3229-3242.
Kuang, S., Charge, S. B., Seale, P., Huh, M. and Rudnicki, M. A. (2006). Distinct roles for Pax7 and Pax3 in adult regenerative myogenesis. *J. Cell Biol.* **172**, 103-113.
Kulesa, P. M. and Fraser, S. E. (2000). In ovo time-lapse analysis of chick hindbrain neural crest cell migration shows cell interactions during migration to the branchial arches. *Development* **127**, 1161-1172.
Kuratani, S. (1997). Spatial distribution of postotic crest cells defines the head/trunk interface of the vertebrate body: embryological interpretation of peripheral nerve morphology and evolution of the vertebrate head. *Anat. Embryol. (Berl.)* **195**, 1-13.
Kuratani, S. (2003). Evolutionary developmental biology and vertebrate head segmentation: a perspective from developmental constraint. *Theor. Biosci.* **122**, 230-251.
Kuratani, S. (2005). Craniofacial development and the evolution of the vertebrates: the old problems on a new background. *Zool. Sci.* **22**, 1-19.
Kuratani, S. (2008a). Evolutionary developmental studies of cyclostomes and the origin of the vertebrate neck. *Dev. Growth Differ.* **50 Suppl. 1**, S189-S194.
Kuratani, S. (2008b). Is the vertebrate head segmented?-evolutionary and developmental considerations. *Integr. Comp. Biol.* **48**, 647-657.
Kuratani, S., Horigome, N. and Hirano, S. (1999). Developmental morphology of the head mesoderm and reevaluation of segmental theories of the vertebrate head: evidence from embryos of an agnathan vertebrate, Lampetra japonica. *Dev. Biol.* **210**, 381-400.
Kusakabe, R. and Kuratani, S. (2005). Evolution and developmental patterning of the vertebrate skeletal muscles: perspectives from the lamprey. *Dev. Dyn.* **234**, 824-834.
Kusakabe, R. and Kuratani, S. (2007). Evolutionary perspectives from development of mesodermal components in the lamprey. *Dev. Dyn.* **236**, 2410-2420.
Kusakabe, R., Kuraku, S. and Kuratani, S. (2011). Expression and interaction of muscle-related genes in the lamprey imply the evolutionary scenario for vertebrate skeletal muscle, in association with the acquisition of the neck and fins. *Dev. Biol.* **350**, 217-227.
Kwon, C., Qian, L., Cheng, P., Nigam, V., Arnold, J. and Srivastava, D. (2009). A regulatory pathway involving Notch1/beta-catenin/Isl1 determines cardiac progenitor cell fate. *Nat. Cell Biol.* **11**, 951-957.
L'Honore, A., Ouimette, J. F., Lavertu-Jolin, M. and Drouin, J. (2010). Pitx2 defines alternate pathways acting through MyoD during limb and somitic myogenesis. *Development* **137**, 3847-3856.
Larroux, C., Luke, G. N., Koopman, P., Rokhsar, D. S., Shimeld, S. M. and Degnan, B. M. (2008). Genesis and expansion of metazoan transcription factor gene classes. *Mol. Biol. Evol.* **25**, 980-996.
Lee, G. H., Chang, M. Y., Hsu, C. H. and Chen, Y. H. (2011). Essential roles of basic helix-loop-helix transcription factors, Capsulin and Musculin, during craniofacial myogenesis of zebrafish. *Cell Mol. Life Sci*. (in press).
Lepper, C., Conway, S. J. and Fan, C. M. (2009). Adult satellite cells and embryonic muscle progenitors have distinct genetic requirements. *Nature* **460**, 627-631.
Lescroart, F., Kelly, R. G., Le Garrec, J. F., Nicolas, J. F., Meilhac, S. M. and Buckingham, M. (2010). Clonal analysis reveals common lineage relationships between head muscles and second heart field derivatives in the mouse embryo. *Development* **137**, 3269-3279.
Lin, C. Y., Yung, R. F., Lee, H. C., Chen, W. T., Chen, Y. H. and Tsai, H. J. (2006). Myogenic regulatory factors Myf5 and Myod function distinctly during craniofacial myogenesis of zebrafish. *Dev. Biol.* **299**, 594-608.
Lin, C. Y., Chen, W. T., Lee, H. C., Yang, P. H., Yang, H. J. and Tsai, H. J. (2009). The transcription factor Six1a plays an essential role in the craniofacial myogenesis of zebrafish. *Dev. Biol.* **331**, 152-166.
Lu, J. R., Bassel-Duby, R., Hawkins, A., Chang, P., Valdez, R., Wu, H., Gan, L., Shelton, J. M., Richardson, J. A. and Olson, E. N. (2002). Control of facial muscle development by MyoR and capsulin. *Science* **298**, 2378-2381.
Mahadevan, N. R., Horton, A. C. and Gibson-Brown, J. J. (2004). Developmental expression of the amphioxus Tbx1/10 gene illuminates the evolution of vertebrate branchial arches and sclerotome. *Dev. Genes Evol.* **214**, 559-566.
Mankoo, B. S., Collins, N. S., Ashby, P., Grigorieva, E., Pevny, L. H., Candia, A., Wright, C. V., Rigby, P. W. and Pachnis, V. (1999). Mox2 is a component of the genetic hierarchy controlling limb muscle development. *Nature* **400**, 69-73.
Mankoo, B. S., Skuntz, S., Harrigan, I., Grigorieva, E., Candia, A., Wright, C. V., Arnheiter, H. and Pachnis, V. (2003). The concerted action of Meox homeobox genes is required upstream of genetic pathways essential for the formation, patterning and differentiation of somites. *Development* **130**, 4655-4664.
Mann, T., Bodmer, R. and Pandur, P. (2009). The Drosophila homolog of vertebrate Islet1 is a key component in early cardiogenesis. *Development* **136**, 317-326.
Maroto, M. and Pourquie, O. (2001). A molecular clock involved in somite segmentation. *Curr. Top. Dev. Biol.* **51**, 221-248.
Matsuoka, T., Ahlberg, P. E., Kessaris, N., Iannarelli, P., Dennehy, U., Richardson, W. D., McMahon, A. P. and Koentges, G. (2005). Neural crest origins of the neck and shoulder. *Nature* **436**, 347-355.
Matt, N., Ghyselinck, N. B., Pellerin, I. and Dupe, V. (2008). Impairing retinoic acid signalling in the neural crest cells is sufficient to alter entire eye morphogenesis. *Dev. Biol.* **320**, 140-148.
McKenzie, J. (1962). The development of the sternomastoid and trapezius muscles. *Cont. Embryol.* **37**, 121-129.
Meadows, E., Cho, J. H., Flynn, J. M. and Klein, W. H. (2008). Myogenin regulates a distinct genetic program in adult muscle stem cells. *Dev. Biol.* **322**, 406-414.

Messina, G., Biressi, S., Monteverde, S., Magli, A., Cassano, M., Perani, L., Roncaglia, E., Tagliafico, E., Starnes, L., Campbell, C. E. et al. (2010). Nfix regulates fetal-specific transcription in developing skeletal muscle. *Cell* **140**, 554-566.
Mjaatvedt, C. H., Nakaoka, T., Moreno-Rodriguez, R., Norris, R. A., Kern, M. J., Eisenberg, C. A., Turner, D. and Markwald, R. R. (2001). The outflow tract of the heart is recruited from a novel heart-forming field. *Dev. Biol.* **238**, 97-109.
Münsterberg, A. E., Kitajewski, J., Bumcrot, D. A., McMahon, A. P. and Lassar, A. B. (1995). Combinatorial signaling by Sonic hedgehog and Wnt family members induces myogenic bHLH gene expression in the somite. *Genes Dev.* **9**, 2911-2922.
Nabeshima, Y., Hanaoka, K., Hayasaka, M., Esumi, E., Li, S., Nonaka, I. and Nabeshima, Y. (1993). Myogenin gene disruption results in perinatal lethality because of severe muscle defect. *Nature* **364**, 532-535.
Nathan, E., Monovich, A., Tirosh-Finkel, L., Harrelson, Z., Rousso, T., Rinon, A., Harel, I., Evans, S. M. and Tzahor, E. (2008). The contribution of Islet1-expressing splanchnic mesoderm cells to distinct branchiomeric muscles reveals significant heterogeneity in head muscle development. *Development* **135**, 647-657.
Neal, H. V. (1918). The history of the eye muscles. *J. Morphol.* **30**, 433-453.
Neyt, C., Jagla, K., Thisse, C., Thisse, B., Haines, L. and Currie, P. D. (2000). Evolutionary origins of vertebrate appendicular muscle. *Nature* **408**, 82-86.
Noden, D. M. (1983a). The role of the neural crest in patterning of avian cranial skeletal, connective, and muscle tissues. *Dev. Biol.* **96**, 144-165.
Noden, D. M. (1983b). The embryonic origins of avian cephalic and cervical muscles and associated connective tissues. *Am. J. Anat.* **168**, 257-276.
Noden, D. M. and Trainor, P. A. (2005). Relations and interactions between cranial mesoderm and neural crest populations. *J. Anat.* **207**, 575-601.
Noden, D. M. and Francis-West, P. (2006). The differentiation and morphogenesis of craniofacial muscles. *Dev. Dyn.* **235**, 1194-1218.
Noden, D. M., Marcucio, R., Borycki, A. G. and Emerson, C. P., Jr (1999). Differentiation of avian craniofacial muscles: I. Patterns of early regulatory gene expression and myosin heavy chain synthesis. *Dev. Dyn.* **216**, 96-112.
Northcutt, R. G. and Gans, C. (1983). The genesis of neural crest and epidermal placodes: a reinterpretation of vertebrate origins. *Q. Rev. Biol.* **58**, 1-28.
Nowotschin, S., Liao, J., Gage, P. J., Epstein, J. A., Campione, M. and Morrow, B. E. (2006). Tbx1 affects asymmetric cardiac morphogenesis by regulating Pitx2 in the secondary heart field. *Development* **133**, 1565-1573.
Okkema, P. G. and Fire, A. (1994). The Caenorhabditis elegans NK-2 class homeoprotein CEH-22 is involved in combinatorial activation of gene expression in pharyngeal muscle. *Development* **120**, 2175-2186.
Olsson, L., Falck, P., Lopez, K., Cobb, J. and Hanken, J. (2001). Cranial neural crest cells contribute to connective tissue in cranial muscles in the anuran amphibian, Bombina orientalis. *Dev. Biol.* **237**, 354-367.
Oustanina, S., Hause, G. and Braun, T. (2004). Pax7 directs postnatal renewal and propagation of myogenic satellite cells but not their specification. *EMBO J.* **23**, 3430-3439.
Palmeirim, I., Henrique, D., Ish-Horowicz, D. and Pourquie, O. (1997). Avian hairy gene expression identifies a molecular clock linked to vertebrate segmentation and somitogenesis. *Cell* **91**, 639-648.
Papaioannou, V. E. and Silver, L. M. (1998). The T-box gene family. *BioEssays* **20**, 9-19.
Pourquié, O., Fan, C.-M., Coltey, M., Hirsinger, E., Wanatabe, Y., Bréant, C., Francis-West, P., Brickell, P., Tessier-Lavigne, M. and Le Douarin, N. (1996). Lateral and axial signals involved in avian somite patterning: a role for BMP4. *Cell* **84**, 461-471.
Relaix, F. and Buckingham, M. (1999). From insect eye to vertebrate muscle: redeployment of a regulatory network. *Genes Dev.* **13**, 3171-3178.
Relaix, F., Rocancourt, D., Mansouri, A. and Buckingham, M. (2005). A Pax3/Pax7-dependent population of skeletal muscle progenitor cells. *Nature* **435**, 948-953.
Relaix, F., Montarras, D., Zaffran, S., Gayraud-Morel, B., Rocancourt, D., Tajbakhsh, S., Mansouri, A., Cumano, A. and Buckingham, M. (2006). Pax3 and Pax7 have distinct and overlapping functions in adult muscle progenitor cells. *J. Cell Biol.* **172**, 91-102.
Rinon, A., Lazar, S., Marshall, H., Buchmann-Moller, S., Neufeld, A., Elhanany-Tamir, H., Taketo, M. M., Sommer, L., Krumlauf, R. and Tzahor, E. (2007). Cranial neural crest cells regulate head muscle patterning and differentiation during vertebrate embryogenesis. *Development* **134**, 3065-3075.
Robb, L., Mifsud, L., Hartley, L., Biben, C., Copeland, N. G., Gilbert, D. J., Jenkins, N. A. and Harvey, R. P. (1998). epicardin: a novel basic helix-loop-helix transcription factor gene expressed in epicardium, branchial arch myoblasts, and mesenchyme of developing lung, gut, kidney, and gonads. *Dev. Dyn.* **213**, 105-113.
Rudnicki, M. A., Schneglesberg, P. N. J., Stead, R. H., Braun, T., Arnold, H.-H. and Jaenisch, R. (1993). MyoD or myf-5 is required for the formation of skeletal muscle. *Cell* **75**, 1351-1359.
Ruppert, E. E. (1997). Cephalochordata (Acrania). In *Microscopic Anatomy of Invertebrates In Hemichordata, Chaetognatha and the Invertebrate Chordates*, vol. 15 (ed. F. W. Harrison and E. E. Ruppert), pp. 349-504. New York: Wiley-Liss.
Rushbrook, J. I., Weiss, C., Ko, K., Feuerman, M. H., Carleton, S., Ing, A. and Jacoby, J. (1994). Identification of alpha-cardiac myosin heavy chain mRNA and protein in extraocular muscle of the adult rabbit. *J. Muscle Res. Cell Motil.* **15**, 505-515.
Sambasivan, R. and Tajbakhsh, S. (2007). Skeletal muscle stem cell birth and properties. *Semin. Cell Dev. Biol.* **18**, 870-882.
Sambasivan, R., Gayraud-Morel, B., Dumas, G., Cimper, C., Paisant, S., Kelly, R. G. and Tajbakhsh, S. (2009). Distinct regulatory cascades govern extraocular and pharyngeal arch muscle progenitor cell fates. *Dev. Cell* **16**, 810-821.
Sauka-Spengler, T., Le Mentec, C., Lepage, M. and Mazan, S. (2002). Embryonic expression of Tbx1, a DiGeorge syndrome candidate gene, in the lamprey Lampetra fluviatilis. *Gene Expr. Patterns* **2**, 99-103.
Schilling, T. F. and Kimmel, C. B. (1997). Musculoskeletal patterning in the pharyngeal segments of the zebrafish embryo. *Development* **124**, 2945-2960.
Schnapp, E., Pistocchi, A. S., Karampetsou, E., Foglia, E., Lamia, C. L., Cotelli, F. and Cossu, G. (2009). Induced early expression of mrf4 but not myog rescues myogenesis in the myod/myf5 double-morphant zebrafish embryo. *J. Cell Sci.* **122**, 481-488.
Seale, P., Sabourin, L. A., Girgis-Gabardo, A., Mansouri, A., Gruss, P. and Rudnicki, M. A. (2000). Pax7 is required for the specification of myogenic satellite cells. *Cell* **102**, 777-786.
Seifert, R., Jacob, M. and Jacob, H. J. (1993). The avian prechordal head region: a morphological study. *J. Anat.* **183**, 75-89.
Shih, H. P., Gross, M. K. and Kioussi, C. (2007). Cranial muscle defects of Pitx2 mutants result from specification defects in the first branchial arch. *Proc. Natl. Acad. Sci. USA* **104**, 5907-5912.
Smith, P. A. and Mango, S. E. (2007). Role of T-box gene tbx-2 for anterior foregut muscle development in C. elegans. *Dev. Biol.* **302**, 25-39.
Stanley, E. G., Biben, C., Elefanty, A., Barnett, L., Koentgen, F., Robb, L. and Harvey, R. P. (2002). Efficient Cre-mediated deletion in cardiac progenitor cells conferred by a 3′UTR-ires-Cre allele of the homeobox gene Nkx2-5. *Int. J. Dev. Biol.* **46**, 431-439.
Starck, D. (1978). Vergleichende Anatomie der Wirbeltiere auf evolutions biologische Grundlage. Berlin: Springer.
Stellabotte, F., Dobbs-McAuliffe, B., Fernandez, D. A., Feng, X. and Devoto, S. H. (2007). Dynamic somite cell rearrangements lead to distinct waves of myotome growth. *Development* **134**, 1253-1257.
Stern, H. M. and Hauschka, S. D. (1995). Neural tube and notochord promote in vitro myogenesis in single somite explants. *Dev. Biol.* **167**, 87-103.
Stockdale, F. E. (1992). Myogenic cell lineages. *Dev. Biol.* **154**, 284-298.
Stolfi, A., Gainous, T. B., Young, J. J., Mori, A., Levine, M. and Christiaen, L. (2010). Early chordate origins of the vertebrate second heart field. *Science* **329**, 565-568.
Tajbakhsh, S. (2009). Skeletal muscle stem cells in developmental versus regenerative myogenesis. *J. Intern. Med.* **266**, 372-389.
Tajbakhsh, S. and Buckingham, M. (2000). The birth of muscle progenitor cells in the mouse: spatiotemporal considerations. *Curr. Top. Dev. Biol.* **48**, 225-268.
Tajbakhsh, S., Rocancourt, D., Cossu, G. and Buckingham, M. (1997). Redefining the genetic hierarchies controlling skeletal myogenesis: *Pax-3* and *Myf-5* act upstream of *MyoD*. *Cell* **89**, 127-138.
Tajbakhsh, S., Vivarelli, E., Kelly, R., Papkoff, J., Duprez, D., Buckingham, M. and Cossu, G. (1998). Differential activation of Myf5 and MyoD by different Wnts in explants of mouse paraxial mesoderm and the later activation of myogenesis in the absence of Myf5. *Development* **125**, 4155-4162.
Theis, S., Patel, K., Valasek, P., Otto, A., Pu, Q., Harel, I., Tzahor, E., Tajbakhsh, S., Christ, B. and Huang, R. (2010). The occipital lateral plate mesoderm is a novel source for vertebrate neck musculature. *Development* **137**, 2961-2971.
Tiecke, E., Matsuura, M., Kokubo, N., Kuraku, S., Kusakabe, R., Kuratani, S. and Tanaka, M. (2007). Identification and developmental expression of two Tbx1/10-related genes in the agnathan Lethenteron japonicum. *Dev. Genes Evol.* **217**, 691-697.
Tirosh-Finkel, L., Elhanany, H., Rinon, A. and Tzahor, E. (2006). Mesoderm progenitor cells of common origin contribute to the head musculature and the cardiac outflow tract. *Development* **133**, 1943-1953.
Tirosh-Finkel, L., Zeisel, A., Brodt-Ivenshitz, M., Shamai, A., Yao, Z., Seger, R., Domany, E. and Tzahor, E. (2010). BMP-mediated inhibition of FGF signaling promotes cardiomyocyte differentiation of anterior heart field progenitors. *Development* **137**, 2989-3000.
Trainor, P. A. and Tam, P. P. (1995). Cranial paraxial mesoderm and neural crest cells of the mouse embryo: co-distribution in the craniofacial mesenchyme but distinct segregation in branchial arches. *Development* **121**, 2569-2582.
Tumer, Z. and Bach-Holm, D. (2009). Axenfeld-Rieger syndrome and spectrum of PITX2 and FOXC1 mutations. *Eur. J. Hum. Genet.* **17**, 1527-1539.
Tzahor, E. (2009). Heart and craniofacial muscle development: a new developmental theme of distinct myogenic fields. *Dev. Biol.* **327**, 273-279.
Tzahor, E. and Lassar, A. B. (2001). Wnt signals from the neural tube block ectopic cardiogenesis. *Genes Dev.* **15**, 255-260.
Tzahor, E., Kempf, H., Mootoosamy, R. C., Poon, A. C., Abzhanov, A., Tabin, C. J., Dietrich, S. and Lassar, A. B. (2003). Antagonists of Wnt and BMP signaling promote the formation of vertebrate head muscle. *Genes Dev.* **17**, 3087-3099.

van Wijhe, J. W. (1882). Über die Mesodermsegmente und die Entwicklung der Nerven des Selachierkopfes. *Ver. Akad. Wiss.* **22**, 1-50.
Verzi, M. P., McCulley, D. J., De Val, S., Dodou, E. and Black, B. L. (2005). The right ventricle, outflow tract, and ventricular septum comprise a restricted expression domain within the secondary/anterior heart field. *Dev. Biol.* **287**, 134-145.
von Scheven, G., Alvares, L. E., Mootoosamy, R. C. and Dietrich, S. (2006a). Neural tube derived signals and Fgf8 act antagonistically to specify eye versus mandibular arch muscles. *Development* **133**, 2731-2745.
von Scheven, G., Bothe, I., Ahmed, M. U., Alvares, L. E. and Dietrich, S. (2006b). Protein and genomic organisation of vertebrate MyoR and Capsulin genes and their expression during avian development. *Gene Expr. Patterns* **6**, 383-393.
Wachtler, F., Jacob, H. J., Jacob, M. and Christ, B. (1984). The extrinsic ocular muscles in birds are derived from the precordal plate. *Naturwissenschaften* **71**, 379-380.
Waldo, K. L., Kumiski, D. H., Wallis, K. T., Stadt, H. A., Hutson, M. R., Platt, D. H. and Kirby, M. L. (2001). Conotruncal myocardium arises from a secondary heart field. *Development* **128**, 3179-3188.
Washington Smoak, I., Byrd, N. A., Abu-Issa, R., Goddeeris, M. M., Anderson, R., Morris, J., Yamamura, K., Klingensmith, J. and Meyers, E. N. (2005). Sonic hedgehog is required for cardiac outflow tract and neural crest cell development. *Dev. Biol.* **283**, 357-372.
Wedin, B. (1949). The anterior mesoblast in some lower vertebrates: a comparative study of the ontogenetic development of the anterior mesoblast in Petromyzon, Etmopterus, Torpedo et al.,. Lund: Hakan Ohlssons Boktryckeri.
Weintraub, H., Davis, R., Tapscott, S., Thayer, M., Krause, M., Benezra, R., Blackwell, T. K., Turner, D., Rupp, R., Hollenberg, S. et al. (1991). The MyoD gene family: nodal point during specification of the muscle cell lineage. *Science* **251**, 761-766.
Yasui, K., Zhang, S., Uemura, M. and Saiga, H. (2000). Left-right asymmetric expression of BbPtx, a Ptx-related gene, in a lancelet species and the developmental left-sidedness in deuterostomes. *Development* **127**, 187-195.
Yu, J. K., Satou, Y., Holland, N. D., Shin, I. T., Kohara, Y., Satoh, N., Bronner-Fraser, M. and Holland, L. Z. (2007). Axial patterning in cephalochordates and the evolution of the organizer. *Nature* **445**, 613-617.
Zacharias, A. L., Lewandoski, M., Rudnicki, M. A. and Gage, P. J. (2011). Pitx2 is an upstream activator of extraocular myogenesis and survival. *Dev. Biol.* **349**, 395-405.