Skeletal muscle is a heterogeneous tissue composed of individual muscle fibers, diversified in size, shape, and contractile protein content, to fulfill the different functional needs of the vertebrate body. This heterogeneity derives from and depends at least in part on distinct classes of myogenic progenitors; i.e., embryonic and fetal myoblasts and satellite cells whose origin and lineage relationship have been elusive so far. In this issue of Genes & Development, Hutcheson and colleagues (pp. 997–1013) provide a first answer to this question.

The origin of embryonic and fetal myoblasts: a role of Pax3 and Pax7

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In the last 10 years, research on muscle progenitors has focused on the two closely related paired domain homeobox transcription factors: Pax3 and Pax7. Their role in different aspects of myogenesis has been widely studied: Pax3 is required for myogenic specification upstream of MyoD (Tajbakhsh et al. 1997), somite segmentation, dermomyotome formation (Tajbakhsh and Buckingham 2000; Schubert et al. 2001; Relaix et al. 2004), limb musculature development (Franz 1993; Bober et al. 1994; Goulding et al. 1994; Relaix et al. 2004), and MyoD and Myf5 expression (Maroto et al. 1997; Bajard et al. 2006), whereas Pax7 is necessary for the maintenance of adult satellite cells (Seale et al. 2000; Oustanina et al. 2004; Relaix et al. 2006). Moreover, the continued growth of muscles that occurs during prenatal and postnatal life has been attributed recently to a population of muscle progenitors already present at the embryonic stage (Gros et al. 2005; Kassar-Duchossoy et al. 2005; Relaix et al. 2005; Schienda et al. 2006). These skeletal muscle progenitor cells arise in the central part of the dermomyotome, coexpress Pax3 and Pax7, can differentiate into skeletal muscle fibers during embryogenesis, and are present as a reserve cell population within the growing muscle mass during prenatal and postnatal life.

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**Pax3/Pax7** double knockout mice, only the primary myotome forms, while all the subsequent phases of myogenesis are compromised; it has therefore been proposed that all the cells of the myogenic lineage may derive from this Pax3/Pax7-positive population of myogenic progenitors. The fate of the Pax3+ cells was followed by using Pax3GFP mice in which the GFP is initially expressed in Pax3-expressing cells; however, because of its stability, GFP expression continues beyond the transient expression of the Pax3 gene product (Relaix et al. 2005). In conclusion, it appears that Pax3 and Pax7 have partially overlapping and partially unique functions in myogenic progenitors; they are both down-regulated during myogenic differentiation, following myogenic regulatory factor (MRF) expression. Despite this abundant information, the specific role that Pax3 and Pax7 play in the specification of embryonic, fetal, and adult myoblasts remained to be investigated. In this issue of *Genes & Development*, Hutcheson et al. (2009) provide a first answer to this question and, based on previous observations (Kardon et al. 2002), also investigate whether Pax3 and Pax7 progenitors also give rise to other cell types.

**Genetic labeling and ablation of myogenic progenitors**

Hutcheson et al. (2009) used genetic lineage tracing and ablation in mice to highlight the developmental origin of embryonic and fetal myoblasts in the limb. By using Pax3Cre and the Pax7iCre mice crossed with the R26RLacZ reporter mice, they confirm a temporal difference in Pax3 and Pax7 expression in line with the idea that Pax3+ and Pax7+ cells could differentially contribute to myogenic lineages. Indeed, several investigators had shown that cells in the epaxial and hypaxial edges of the dermomyotome, and also cells migrating to the limb, express Pax3 but not Pax7 (for review, see Buckingham and Relaix 2007). However, lineage ablation analysis (using the R26RDTA/+ mice) demonstrates the existence of Pax33°Pax7° somitic cells required for embryonic myogenesis and of Pax3°Pax7° cells required for fetal and subsequently adult myogenesis. These Pax7° cells derive from Pax3° cells but no longer express Pax3, as Pax3 is down-regulated after E13.5. Thus, in the limb, embryonic and fetal myoblasts arise from developmentally distinct, although related, progenitors (Fig. 1). Moreover, the lineage analysis revealed that the Pax3+, but not the Pax7+, cells in the limb are bipotential, contributing to both muscle and endothelial cell lineage, suggesting that these Pax3+ cells in the limb retain the properties of their sisters in the hypaxial dermomyotome initially (Kardon et al. 2002). This important result leads to a crucial and yet unanswered question: Although Pax3 is a key regulator of skeletal muscle development (Maroto et al. 1997; Tajbakhsh et al. 1997; Bajard et al. 2006; Buchberger et al. 2007), in vivo expression of this gene in somitic cells is not, per se, sufficient to commit these cells toward a myogenic fate (since also endothelial cells are generated), strongly suggesting that additional extrinsic cues are needed to dictate the choice between myogenic and endothelial differentiation. The role of Pax7 in fetal myogenesis is consistent with previous expression data (Horst et al. 2006; Biressi et al. 2007). Nevertheless, in the Pax7-null mouse, in which adult myogenesis and regeneration are seriously compromised, fetal myogenesis is not (Seale et al. 2000; Oustanina et al. 2004; Relaix et al. 2006). Hutcheson et al. (2009) suggest that during fetal myogenesis, Pax7°’s function either is not essential or is compensated...
by other proteins, the most likely candidate being Pax3. In other words, Pax3 would be essential for embryonic myogenesis and Pax7 for adult myogenesis, whereas during the fetal period the two genes would share redundant functions.

**β-Catenin is required for embryonic but not fetal myogenesis**

The study by Hutcheson et al. [2009] goes on to show a β-catenin requirement for determination, migration, and proliferation of the Pax3/iPax7-expressing cells. Different studies have demonstrated the importance of Wnt/β-catenin signaling for muscle determination [Maroto et al. 1997; Tajbakhsh et al. 1997; Borello et al. 1999, 2006; Chen et al. 2005; Brunelli et al. 2007], and for dermomyotome and myotome formation [Ikeya and Takada 1998; Linker et al. 2003; Schmidt et al. 2004; Otto et al. 2006]. Here, Hutcheson et al. [2009] compare the effects of β-catenin deletion or activation driven by either Pax3Cre or Pax7Cre to test the role of Wnt/β-catenin signaling in embryonic and fetal myoblast differentiation. Results show that β-catenin is necessary for dermomyotome development, as predictable from previous work; once in the limb, however, β-catenin is no longer required for embryonic myoblasts, while Pax7 fetal myoblasts [and the fibers they form] are reduced in number in the absence of β-catenin signaling, suggesting a role in mediating proliferation signals. It should be noted that the work has been carried out on limb myoblasts: Although a similar scenario may exist of myoblasts in the trunk, this remains to be demonstrated.

**Questions for the future**

The study by Hutcheson et al. [2009] adds an important piece to the complex and still confusing puzzle of muscle development by defining the role of Pax3/7 genes and β-catenin in embryonic and fetal myoblasts [Fig. 1]. Several open questions remain: What is the fate of the Pax3β-Catenin -derived, Myf5 embryonic myoblasts? Do they all differentiate, or rather contribute to later (fetal and adult) myogenic cells? Do all satellite cells derive from Pax3/7 embryonic/fetal myoblasts? Could other cell types feed into this compartment and be recognized as part of the myoblast population simply because they activate Pax3 and/or Pax7? Clearly, more markers and more lineage tracing experiments are needed to answer these questions. Equally important will be the identification of new molecules or molecular pathways downstream from Pax3 and/or Pax7 and responsible for myoblast diversification. In this perspective, it would be important to understand whether Pax3 alone is sufficient to rescue fetal myogenesis in Pax7-null mice or whether yet-to-be-identified additional molecules may cooperate with it in this role. New data will answer these questions in a hopefully near future.

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**References**


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