whereas in cereals and grasses, they do not seem to be present, with the exception of the Avena (oat) genus. A series of DNA and RNA blot analyses in several cereals did not identify any orthologs of the avenacin Sad1 gene (Qi et al., 2004). Of note, oat possibly diverged from the eudicots about 150 million years ago. It has been demonstrated that extensive gene loss of secondary metabolite synthases in microorganisms could mistake a case of common ancestry for horizontal gene transfer or convergent evolution (Rantala et al., 2004). However, phylogenetic analyses of the avenacin and thalianol pathways show that the genes of each pathway are monocot and eudicot specific, respectively. Therefore, Field and Osbourn (2008) exclude horizontal gene transfer from bacteria or other organisms. Unless we are dealing with a case of rampant gene duplication, rapid neofunctionalization, and gene losses of the ancestral genes, which actually might not be so far-fetched for secondary metabolism, the most reasonable scenario is convergent evolution and repeated de novo synthesis of the avenacin and thalianol gene clusters, as suggested by the authors. Cases of convergent evolution have been reported before for regulatory networks involved in animal development (Amoutzias et al., 2004) or for the formation of pathways of secondary metabolism (Pichersky and Gang, 2000). Nevertheless, the birth of clusters of functionally related genes with tightly coordinated expression seems a rather rare event in eukaryotic genomes, but, undoubtedly, upcoming plant genomes will shed more light on fundamental questions regarding the structure and evolution of such gene clusters (see for example Figure 1).

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Glucose Restriction: Longevity SIRTainly, but without Building Muscle?

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The two metabolic sensors AMPK and SIRT1 take center stage as Fulco et al. reveal, in this issue of *Developmental Cell*, the signaling mechanism by which low glucose prevents the correct development of the myogenic program. These observations may hold some therapeutic promise against muscle wasting.

One of the most amazing features of skeletal muscle is its high plasticity, enabling it to respond to changes in activity, injury, or degeneration. This plasticity is largely due to muscle stem cells, better known as satellite cells, residing beneath the basal lamina of adult skeletal muscle, closely juxtaposed against the muscle fibers (Le Grand and Rudnicki, 2007), which have the ability to modulate muscle growth and differentiation. Satellite cells in adult skeletal muscle are normally quiescent, but proliferation and differentiation of their descendant cells can be activated by diverse forms of stress, thereby playing an essential role in muscle regeneration, muscle hypertrophy, and postnatal muscle growth.

Despite the astonishing advance during the last few decades in our understanding of the process of myogenesis (reviewed in Le Grand and Rudnicki, 2007), the molecular mechanisms regulating the differentiation of myogenic stem cells are still unclear. In this issue of *Developmental Cell*, a new report from Fulco et al. (2008) examines the effects of nutrient availability on myogenic differentiation. Interestingly, the authors find that restricted glucose availability prevents myogenesis. Furthermore, the authors identify AMP-activated protein kinase (AMPK)—a master switch of anabolic versus catabolic processes—as a key sensor of low glucose levels during myogenesis. The concept that low glucose levels leads to the

activation of AMPK is not new, but it is the first time that this enzyme is shown to be directly involved in myogenesis. The inhibitory role of AMPK on myocyte differentiation is highlighted by results showing that myotubes differentiate normally in glucose-restricted conditions when AMPK is inhibited. Conversely, artificial activation of AMPK with 5-aminoimidazole-4-carboxamide-1-{beta}-D-ribofuranoside (AICAR), the standard compound used to activate AMPK, is enough to prevent myogenesis even in conditions when glucose is not limiting. It is interesting to note that AMPK has already been shown to inhibit differentiation in other cell models, such as adipocytes (Habinowski and Witters, 2001). This new role of AMPK would increase the already wide spectrum of functions of this pleiotropic enzyme.

Perhaps the most relevant and unexpected piece of data provided by Fulco et al. might be the mechanism by which AMPK activation inhibits muscle differentiation. The authors find that AMPK activation causes an increase in the cellular NAD^+/NADH ratio, which in turn is sensed by the NAD^+-dependent type III histone deacetylase SIRT1. In a previous study, the authors have already implicated SIRT1 as a negative regulator of myogenesis (Fulco et al., 2003). When NAD^+ levels increase, SIRT1 deacetylates and inactivates MyoD, a key transcriptional regulator of myogenesis, thereby preventing the myogenic program (Fulco et al., 2003). Therefore, the induction of the deacetylase activity of SIRT1 seems to be the vehicle through which AMPK blocks myocyte differentiation. Supporting this hypothesis, AMPK fails to prevent myogenesis in myocytes where SIRT1 expression has been inhibited with specific short hairpin RNAs.

As happens with most important discoveries, the findings by Fulco et al. open the door for more questions. Further investigation will have to establish whether the modulation of AMPK or SIRT1 activity is relevant during situations where the organism has to build muscle "in vivo." For this purpose, it would be interesting to test whether any of the diverse transgenic mice models for either AMPK or SIRT1 display abnormal muscle development and whether their response to muscle regeneration or hypertrophic stimuli, such as insulin-like growth factors or resistance exercise, is compromised. Furthermore, the effects of these metabolic sensors on myogenesis could also be effectively probed through the use of specific pharmacological agonists that target either AMPK or SIRT1 (Cool et al., 2006; Milne et al., 2007). Moreover, other players that could participate to mediate the effects of glucose deprivation on myogenesis should be explored. For example, is AMPK inhibiting myogenesis solely through SIRT1 or are other AMPK targets, like the mTOR pathway or the FOXO family of transcription factors, involved? In this context, it is relevant that both the inhibition of mTOR (Cuenda and Cohen, 1999) and the activation of FOXO (Hribal et al., 2003) have been shown to negatively regulate myogenesis. Likewise, can all the effects be attributed to a negative regulation of MyoD or are there other deacetylation substrates of SIRT1 that regulate myogenesis?

How AMPK activition modulates SIRT1 activity and whether the alterations of the NAD^+/NADH ratio are responsible for SIRT1 activation is also particularly important to establish. The authors propose that AMPK increases the expression of the nicotinamide phosphoribosyltransferase (Nampt), an enzyme that catalyzes NAD^+ synthesis from nicotinamide (Revollo et al., 2004). In line with this hypothesis, knocking down Nampt prevents the effects of AMPK on the NAD^+/NADH ratio and, consequently, on SIRT1 activation. However, these conclusions are based on studies performed 36–48 hr after the pharmacological activation of AMPK, which may be too long for a conclusive readout of enzymatic activity changes. Therefore, it could be worthwhile to explore whether the direct regulation of Nampt enzymatic activity by AMPK, and not only its expression, could contribute to the observed effects. Post-translational modifications such as phosphorylation, and perhaps ubiquitination or acetylation, could be relevant in this context.

Altogether, this interesting study sheds a new light on our understanding of myogenesis and shows the importance of nutrient-gated pathways controlled by AMPK and SIRT1 in muscle formation. AMPK and SIRT1, two of today's prime targets for the treatment of metabolic and age-related disease, now also become endowed with a novel and promising role, with potential therapeutic implication against the devastating effects of muscle wasting. We furthermore predict that the link between AMPK and SIRT1 signaling will become a fertile ground for future investigations, because it could have an impact beyond myogenesis (Figure 1). In fact, the modulation of SIRT1 activity by AMPK would tie these metabolic sensors together and perhaps explain not only why treatment with SIRT1 activators (Lagouge et al., 2006; Milne et al., 2007) mimics many of the effects associated with AMPK activation (Cool et al., 2006), but also why SIRT1 and AMPK have similar effects on metabolism and lifespan.

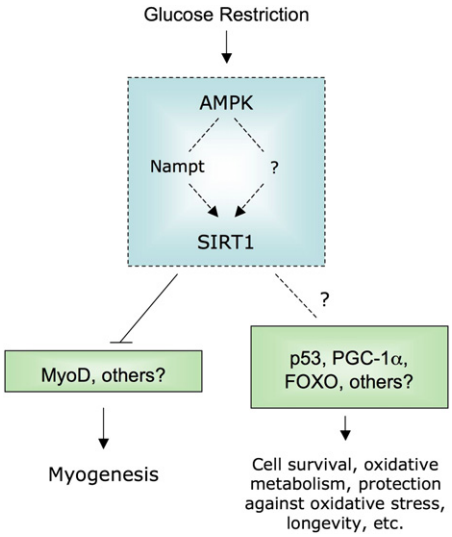

Figure 1. Possible Pleiotropic Actions of Glucose Restriction through AMPK and SIRT1
SIRT1 can deacetylate and inhibit MyoD, a key transcription factor for myogenesis. Activation of AMPK during glucose restriction triggers the activation of SIRT1, thereby blocking the induction of the myogenic program. However, triggering SIRT1 activity by AMPK could also translate into the regulation of other SIRT1 deacetylation targets, such as p53, peroxisome-proliferator activated receptor gamma coactivator 1α (PGC-1α), or the FOXO family of transcription factors, leading to a plethora of possible effects that perhaps extend beyond the regulation of myogenesis.

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Cyclins in Meiosis: Lost in Translation

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In a recent issue of *Cell*, Carlile and Amon examine the regulation of four budding yeast B-type cyclins, crucial for regulating and distinguishing meoisis I and meoisis II divisions, and find a surprising diversity of behaviors and modes of regulation. In particular, Clb3 is regulated by a striking translational repression specific to meoisis I.

Meiosis is often portrayed as a specialized cell division, conjuring the image of a mitotic ("vegetative") division with bells and whistles. But when one looks in detail at what happens, and why and how, one is struck by the vast differences between the vegetative and the meiotic divisions. Many of the same proteins and processes are used, yes, but the relationships among them, and the regulatory wiring, are often unrecognizable.

In a vegetative division, chromosomes are replicated, and then at mitosis the paired sister chromatids segregate from each other, producing two identical cells. This process depends in part on the protein kinase activity of a cyclin-dependent kinase (CDK, Cdc28 in budding yeast) bound to and activated by a B-type cyclin (most importantly Clb2 in budding yeast). Entry into anaphase depends on high levels of CDK activity, and exit from anaphase depends on loss of this activity.

In a meiotic division (the point of which is to generate haploid gametes from a diploid parent), there is likewise replication of chromosomes, but followed by two rounds of division. At meoisis I, one pair of sister chromatids recombines with, then segregates from, its homologous pair (a division quite unlike anything seen in vegetative cells), while at meoisis II, the sister chromatids segregate from each other (a division similar to the vegetative division). For both meoisis I and meoisis II, entry into anaphase depends on high CDK protein kinase activity, and exit from anaphase depends on loss of this activity.

This complicated chromosome dance presents the cell with a number of challenges. First, chromosome behavior in meoisis I and meoisis II must be reliably different. Second, there is a need to coordinate loss of CDK activity to complete meoisis I with gain of CDK activity to initiate meoisis II. This second issue has been well-studied in *Xenopus* (e.g., Furuno et al., 1994; Hochegger et al., 2001).

Fortunately, budding yeast has six B-type cyclins, Clb1 through Clb6 (reviewed by Bloom and Cross, 2007). In principle, these could to some extent direct the Cdc28 cyclin-dependent kinase to phosphorylate different substrates, and furthermore, each cyclin could be independently regulated at various levels. In the vegetative cell cycle, some of these cyclins are regulated by transcription and by protein degradation, and the various cyclin-CDK complexes are differentially sensitive to inhibitors such as Sic1 and perhaps also to regulators such as Swe1 and Mih1 (reviewed by Bloom and Cross, 2007; Mendenhall and Hodge, 1998).

Thus, at least in principle, the differences between the vegetative, meoisis I, and meoisis II divisions could be partly due to differences in the properties and regulation of the six B-type cyclins. Indeed, genetic and other studies have shown that the major vegetative B-type cyclin, Clb2, is not expressed in meiosis (Grandin and Reed, 1993) and has no role in meiotic events. Loss of *CLB1*, *CLB3*, or *CLB4* has distinguishable meiotic phenotypes, though mechanistically it is not clear why (Dahmann and Futcher, 1995).

Carlile and Amon (2008) have addressed these issues by examining the behavior of the mRNAs, proteins, and protein kinase activities of Clb1, Clb3, Clb4, and Clb5 through meiosis. Key to this examination was a new method for producing highly synchronous meiotic cultures, allowing meoisis I and meoisis II to be resolved. They found a truly striking diversity in the patterns and modes of regulation of these related cyclin genes. For all four genes, transcription is upregulated before meoisis I and downregulated after meoisis II, and protein levels diminish with transcript levels, suggesting protein turnover. However, novel, gene-specific patterns of regulation by other mechanisms also appear. With respect