Not Too Large and Not Too Small–Just the Right Size: A Hippo-Sized Heart

Edward E. Morrisey

Hippo Pathway Inhibits Wnt Signaling to Restrain Cardiomyocyte Proliferation and Heart Size
Heallen et al
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Hippo signaling is an important regulator of organ size by balancing both cell proliferation and apoptosis. In cardiac development, Wnt signaling has been shown to be an important mediator of early cardiomyocyte proliferation and differentiation. Recent studies have now shown that the Hippo pathway negatively regulates heart size through recruitment of the coactivator Yap to the Wnt transcriptional effector β-catenin to balance the level of cardiomyocyte proliferation.[1]

How organs obtain the correct size during development is one of the least understood processes during development. The coordination of the proper level of cell proliferation as well as differentiation and maturation is regulated by multiple developmental pathways, but few have been specifically implicated in controlling organ size. Heart function is quite sensitive to the effects of cardiac organ size, especially during development. A heart that is too small can lead to cardiac insufficiency and developmental demise. In contrast, in the adult, pathological cardiac hypertrophy, which can cause an enlarged heart, can result in decreased cardiac function and severe heart disease. Thus, during development, regulation of cardiac size can have an important impact on cardiovascular function.

The Hippo pathway was originally shown to regulate organ size in Drosophila imaginal disc development.[2,3] In vertebrates, the function of the Hippo pathway is less understood, but several orthologues of the Drosophila Hippo genes Mst1 and Mst2, which are serine/threonine kinases, have been identified as well as other components in the pathway. In a recent report, Heallen et al demonstrate that inactivation of one of these core components, the scaffolding protein Salvador, which interacts with Mst1/2 to form an active kinase

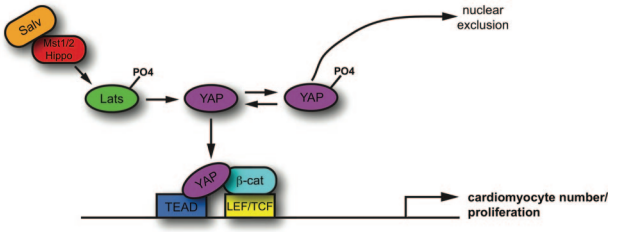

Figure. The Hippo pathway component Salvador (Salv) interacts with the Mst1/2 homologues of the Hippo protein, and this complex regulates phosphorylation of the Lats tumor suppressor gene. Lats in turn regulates phosphorylation of YAP. Phosphorylated YAP is excluded from the nucleus. Nonphosphorylated YAP interacts with both TEAD transcription factors and β-catenin, providing an important connection between the Hippo and Wnt/β-catenin pathways that controls cardiomyocyte number and proliferation.

complex required for Hippo pathway function, leads to a dramatic form of cardiomegaly.[1] The authors show that the increase in heart size was accompanied by an increase in cardiomyocyte mitosis but not by an increase in Isl1+ cardiac progenitor proliferation. The authors went on to uncover an interesting mechanism for Hippo function by showing that the Yap coactivator, which is a transcriptional effector of Hippo signaling, complexes with β-catenin to promote Wnt signaling in developing cardiomyocytes. This coactivation of Wnt/β-catenin signaling leads to the increased expression of a number of interesting target genes including Sox2, survivin/Birc5, and Snail2. Loss of β-catenin in the developing heart leads to decreased expression of these genes, supporting the finding that they are regulated by both Hippo and Wnt/β-catenin signaling. Given the importance of Wnt/β-catenin signaling in promoting cardiomyocyte proliferation and differentiation, these findings present a novel input that helps control cardiomyocyte proliferation to regulate overall heart size during development.

Much of what is known about Wnt/β-catenin signaling in early cardiac development suggests a primary role in regulating proliferation and differentiation in second heart field progenitors.[4–8] However, Heallen et al did not observe any difference in the proliferation rates between second heart field–derived and first heart field–derived structures, for example, right and left ventricular myocardium, respectively. This finding is somewhat unexpected and may suggest that the Hippo pathway interacts with the Wnt/β-catenin pathway in both areas of the heart, but its role in first heart field is different than in the second heart field. One possibility is that components of the Hippo pathway may not be expressed in the second heart field and thus do not effect Wnt signaling. Alternatively, our understanding of Wnt/β-catenin function in

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the first heart field is incomplete and may be more complex than previously described.

One of the conundrums of Wnt/β-catenin signaling in cardiac development has been the observation that several Wnt ligands are expressed in the myocardium much later and more broadly than Wnt/β-catenin signaling has been observed using several methods including LEF/TCF transgenic reporters.[4,7,9-11] Moreover, current concepts in cardiomyocyte differentiation from pluripotent stem cells suggest a biphasic role for Wnt signaling with the pathway having a procardiogenic role in development of precardiac mesoderm and an anticardiogenic role later during cardiomyocyte differentiation.[12,13] One interpretation of these data is that Wnt/β-catenin signaling may have little effect on cardiomyocyte proliferation after early progenitor expansion. However, this may be too simplistic. It is clear in other developmental systems that different Wnt ligands have different effects on Wnt/β-catenin signaling. Noncanonical Wnt pathways that could act independent of β-catenin signaling may play a key role in balancing cardiomyocyte proliferation and differentiation. Noncanonical Wnt signaling can also inhibit canonical β-catenin dependent signaling, leading to a precise level of overall Wnt activity in cells and tissues that express multiple Wnt ligands. The findings by Heallen et al suggest an additional input for Wnt/β-catenin signaling by the Hippo pathway, thus providing another level of control to maintain cardiomyocyte proliferation and ultimately heart size.

How Yap/β-catenin interactions balance out the canonical LEF-TCF/β-catenin interactions and whether these interactions are mutually exclusive or can occur simultaneously are points that still need to be more fully characterized. Only nonphosporylated Yap interacts with β-catenin in this situation and activation of Hippo signaling phosphorylates Yap, leading to nuclear exclusion and inactivation of Yap/β-catenin transcriptional activity. It is conceivable that the interaction between Yap/β-catenin and either TEAD or LEF-TCF factors plays a crucial role in regulating the precise and gradual exit of the cell cycle noted in cardiomyocytes. This may be similar to how β-catenin interacts with some members of the Sox transcription factor family, resulting in inhibition of Wnt/β-catenin signaling.[14,15] Thus, differing inputs into the Wnt/β-catenin signaling pathway may play crucial roles in regulating cardiomyocyte growth during later stages of cardiac development.

Although the full affects of Hippo signaling on cardiac organ size remain to be explored, the present study by Heallen et al strongly implicates that this pathway intersects with Wnt signaling to control cardiac size during development. These studies also outline potentially new targets for therapeutic approaches to modulate cardiomyocyte proliferation. The Mst1/2 kinases may provide attractive new targets for therapeutic inhibition to promote cardiomyocyte proliferation in the setting of injury. Inhibition of Gsk-3β, which regulates Wnt/β-catenin signaling, has been shown to promote cardiomyocyte proliferation during development and in postnatal cardiomyocytes.[16] Whether activation of Wnt or inactivation of Hippo signaling will promote cardiomyocyte proliferation in the setting of cardiac injury is still unknown. However, the current findings are sure to spur interest in looking more closely into these pathways in the adult heart.

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