Does cardiac development provide heart research with novel therapeutic approaches? [version 1; peer review: 2 approved]

Angeliqua Sayed, Mariana Valente, David Sassoon

Cellular, Molecular, and Physiological Mechanisms of Heart Failure, Paris-Cardiovascular Research Center (PARCC), European Georges Pompidou Hospital (HEGP), INSERM U970, F-75737 Paris Cedex 15, Paris, France

First published: 06 Nov 2018, 7(F1000 Faculty Rev):1756
https://doi.org/10.12688/f1000research.15609.1

Abstract
Embryonic heart progenitors arise at specific spatiotemporal periods that contribute to the formation of distinct cardiac structures. In mammals, the embryonic and fetal heart is hypoxic by comparison to the adult heart. In parallel, the cellular metabolism of the cardiac tissue, including progenitors, undergoes a glycolytic to oxidative switch that contributes to cardiac maturation. While oxidative metabolism is energy efficient, the glycolytic-hypoxic state may serve to maintain cardiac progenitor potential. Consistent with this proposal, the adult epicardium has been shown to contain a reservoir of quiescent cardiac progenitors that are activated in response to heart injury and are hypoxic by comparison to adjacent cardiac tissues. In this review, we discuss the development and potential of the adult epicardium and how this knowledge may provide future therapeutic approaches for cardiac repair.

Keywords
hypoxia, glycolytic, oxidative, cardiac progenitor, epicardium

Open Peer Review
Approval Status ✓ ✓

version 1 06 Nov 2018

Faculty Reviews are review articles written by the prestigious Members of Faculty Opinions. The articles are commissioned and peer reviewed before publication to ensure that the final, published version is comprehensive and accessible. The reviewers who approved the final version are listed with their names and affiliations.

1. Jop H. van Berlo, Lillehei Heart Institute and Stem Cell Institute, Minneapolis, USA
2. Robert G. Kelly, University of Aix-Marseilles, CNRS UMR 7288, Marseilles Cedex 9, France

Any comments on the article can be found at the end of the article.
Corresponding author: David Sassoon (david.a.sassoon@gmail.com)

Author roles: Sayed A: Conceptualization, Visualization, Writing – Original Draft Preparation, Writing – Review & Editing; Valente M: Conceptualization, Visualization, Writing – Original Draft Preparation, Writing – Review & Editing; Sassoon D: Conceptualization, Funding Acquisition, Project Administration, Supervision, Validation, Visualization, Writing – Original Draft Preparation, Writing – Review & Editing

Competing interests: No competing interests were disclosed.

Grant information: The laboratory is supported by the Laboratoire d'Excellence Revive (Investissement d'Avenir, ANR-10-LABX-73), the Fondation Leducq (grant 13CVD01; CardioStemNet project), and Agence Nationale pour la Recherche (ANR) grant RHU CARMMA (ANR-15-RHUS-0003). The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Copyright: © 2018 Sayed A et al. This is an open access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

How to cite this article: Sayed A, Valente M and Sassoon D. Does cardiac development provide heart research with novel therapeutic approaches? [version 1; peer review: 2 approved] F1000Research 2018, 7(F1000 Faculty Rev):1756
https://doi.org/10.12688/f1000research.15609.1

First published: 06 Nov 2018, 7(F1000 Faculty Rev):1756 https://doi.org/10.12688/f1000research.15609.1
Introduction

Cardiovascular diseases represent a prevalent medical challenge and are a leading cause of morbidity and mortality in developed societies. Although multiple approaches have been developed in the last two decades to repair or replace damaged myocardium, progress to date has been modest and cardiomyocyte replacement in adult hearts remains elusive. While understanding the loss of cardiac muscle and its eventual replacement is an important goal for cardiac research, there are additional cellular targets that hold promise. These include abrogating scar tissue formation following heart injury or promoting robust revascularization to the injured heart tissue. Here, we present an overview of the different cell types that participate in mammalian heart development as well as how the microenvironment participates in cardiac remodeling. Typically, tissue repair is driven by tissue-resident progenitors. Although significant debate exists regarding the identity and functional potential of adult cardiac progenitor cells (CPCs), there is an emerging consensus that the adult epicardium contains cells with progenitor potential that can participate in cardiac remodeling after injury. Whereas the heart undergoes profound changes during embryonic, fetal, and early postnatal growth, the epicardium maintains many characteristics found during heart development. Consequently, the adult epicardium is an important cellular compartment that may provide solutions to ameliorating heart repair. We focus particularly on rodent models that have allowed the identification of progenitor populations that give rise to the heart as well as for the study of specific cell types in response to injury.

Cardiac progenitor populations during development

The mature heart consists of an inner layer of endocardial cells (endocardium) and an outer layer of epicardial cells (epicardium) that surround the myocardium. The myocardium is composed of cardiomyocytes as well as cells of the conductive system, smooth muscle, and endothelium and stromal cells/fibroblasts and valvular interstitial cells. Three sets of multipotent precursors have been identified that give rise to cardiac cells: cardiogenic mesoderm cells, cardiac neural crest cells (cNCCs), and proepicardial organ (PEO) cells (Figure 1A). Cardiogenic mesoderm cells give rise to cardiomyocytes by progressive lineage restriction that segregate during the onset of gastrulation. During murine development, a first lineage appears at embryonic day 7.0 (E7.0), forms the cardiac crescent (first heart field, or FHF), and is the primary source of the left ventricular cardiomyocytes and primitive atria. While the cardiac tube begins to contract and pump blood throughout the embryo, a second lineage appears at E8.0 that constitutes the second heart field (SHF) and gives rise to the right ventricle, both atria, and the outflow tract (OFT). Unlike the FHF, the SHF consists of multipotent cells that give rise to cardiomyocytes as well as endothelial cells and the smooth muscle of the OFT region. The multipotent capacity of the SHF has been assessed by using embryonic stem cells or in vivo analyses at the population level; thus, the precise nature and clonal behavior of the SHF progenitors warrant further investigation. At the looping heart tube stage, when both FHF and SHF progenitors are in place, myocardium trabeculation occurs, leading to an increase of the endocardial and myocardial surface area that promotes oxygenation of the tissue. The base of the trabeculae contains a population of proliferative cells—referred to as the compact layer—that are essential to ventricular wall expansion and participate in the development of the coronary tree. It has been proposed that the epicardium secretes mitogenic signals that drive cardiomyocyte proliferation in the compact zone (Figures 1A, 1B).

Another cellular compartment derived from cardiogenic mesoderm SHF consists of specialized endothelial cells (endocardium) that cover the inner surface of the cardiac tube and participate in different heart morphogenesis processes, which include trabeculation and a part of the coronary tree formation. These two developmental events are intimately related, as the endocardium is essential to promote trabeculation of the ventricles and participates in the exchange of gas and nutrients with the blood circulating in the ventricular chambers. During septation, ventricular trabeculae initiate compaction, increasing the thickness of the ventricular wall. Endocardial cells surrounding the trabeculae become entrapped in the compacted myocardium and adopt a capillary-like morphology. The newly formed endocardial-derived capillaries subsequently connect with the epicardial-derived vascular network. In addition, the endocardium is an important source of mesenchymal cells for endocardial cushion development and subsequent valvulogenesis and membranous septation. During the septation, endocardial cells undergo an endothelial-to-mesenchymal transition (EndoMT) and colonize the subjacent cardiac jelly at the level of the atrioventricular canal at E9.5 and the OFT at later stages (Figures 1A, 1B).

Cardiac neural crest derived from the dorsal neural tube at E9.5 migrates through the pharyngeal arches and gives rise to smooth muscle and mesenchymal cells that participate in the septation of the OFT trunk (Figure 1A, B).

The PEO is an extra-cardiac transient cauliflower-like structure that appears between E8.5 and E10.5 adjacent to the venous pole of the heart tube (splanchic mesoderm-derived). PEO cells attach to the myocardium surface and spread out to constitute the outer layer of the looping heart tube (that is, epicardium). A subset of epicardial cells undergoes epithelial-to-mesenchymal transition (EMT) in the subepicardial region referred to as mesenchymal epicardium-derived cells (EPDCs). A small fraction of EPDCs invades the myocardium to give rise to stromal cells/intertstitial fibroblasts and coronary vasculature. The contribution of the PEO to cardiomyocytes and endothelial cells has been proposed but remains less clear. An infrequent contribution to cardiomyocytes by the epicardium has been reported to be restricted to specific cardiac zones (inter-ventricular septum and parts of the atrial myocardium). In addition, it has been shown recently that nascent coronary vasculature forms immediately subjacent to the epicardium (subepicardial zone) and contributes to a large proportion of the coronary arteries, veins, and capillaries in the myocardial compact layer. Following the formation of the initial vascular plexus, the coronary vessels connect to the aorta. Irrespective of the origin, the coronary vasculature is essential for the subsequent heart morphogenesis and embryonic viability such that deficient coronary
Figure 1. Patterning of mouse heart development. (A) Hierarchical relationship of the different cardiac developmental progenitors and their progeny. (B) Main developmental steps of heart morphogenesis. A color code was assigned and followed to define each cardiac progenitor population and their progeny. AVN, atrioventricular node; AVRB, atrioventricular ring bundle; CM, cardiomyocyte; E, embryonic day; FHF, first heart field; PVC, peripheral ventricular conduction system; SAN, sinoatrial node; SHF, second heart field.
development impairs myocardium compaction and leads to embryonic lethality37,38,91 (Figure 1A, B). The cardiac conduction system is composed of specialized cardiomyocytes that assemble into a complex and heterogeneous structure to make up a central conduction system located in the atria (sinoatrial node [SAN], atrioventricular ring bundles, and the atrioventricular node) and a peripheral conduction system in the ventricles (left and right branches of His bundle, LBB and RBB, and left and right peripheral conductive system or the Purkinje fibers)96. SAN cardiomyocytes derive from the posterior SHF; however, lineage analyses revealed early lineage segregation from the atrial working cardiomyocyte progenitors56,66,77. Retrospective clonal analyses showed that the nlacZ gene transmitted cells are organized into clusters of clonally related cardiomyocytes, which are composed of a mixture of both working and specialized conductive cardiomyocytes68. These nlacZ clusters were found in all conductive system compartments, except in the SAN, indicating a common origin between the specialized conductive cardiomyocytes and their neighbor working cardiomyocytes66,68,69. Of note, this retrospective clonal analysis does not exclude the contribution of other progenitor populations to the conductive system69. Additionally, other cardiovascular progenitors—that is, PEO (EPDCs)70 and cNCCs71—have been suggested to contribute to the development of the conductive system; however, this participation remains controversial72,73,80,81 (Figures 1A, B).

Our understanding of heart development is that specific progenitors arise at discrete developmental stages originating from different embryonic structures to form the heart. Thus, although the development of a tissue and an understanding of the progenitors involved can shed light on whether CPCs exist in the adult tissue, the heart poses a considerably more complex situation. Indeed, the search for adult CPCs has remained difficult and at times contentious regarding their identity as well as their cell fate potentials. While many tissues are capable of robust regeneration following injury, the mammalian heart shows a limited capacity to repair, suggesting that, if CPCs exist, their capacities are extremely limited. One possible reason for this limited capacity is that the CPC microenvironment of the adult is not permissive and thus the microenvironment of the developing heart warrants further study.

**The developmental cardiac progenitor microenvironment**

Placental pregnant females are commonly exposed to normal levels of oxygen (normoxia, 21% oxygen), and in turn the placenta regulates the level of oxygen available during embryonic and fetal development (Box 1 and Figure 2). The microenvironment of the developing heart is characterized by low levels of oxygen (physiological hypoxia) (Box 2) that promotes glycolytic metabolism72,73. Oxygen tension is an important signal driving tissue development and maturation74–76,78,79. Several studies have shown that physiological hypoxia regulates multiple cellular processes, including stem cell maintenance, proliferation, and differentiation, particularly in the context of angiogenesis and formation of placenta, heart, cartilage, and bone72,74,75,80,81. In addition, the developing cardiovascular system is unable to homogeneously deliver nutrients and oxygen throughout the embryonic/fetal heart61,63,76,77,83. Until mid-gestation, the coronary vasculature is absent and there is a gradient of oxygenation established from the endocardium (more oxygenated and in direct contact with the blood from the chambers) to the epicardium (less oxygenated61,63,83. The myocardial compact layer (epicardial side) is composed of immature cardiomyocytes undergoing higher levels of proliferation under lower oxygen tension as compared with cardiomyocytes in the trabeculae (endocardial side)28,30,83. Ultimately, the coronary vasculature is established to meet the increasing demands of the myocardial wall expansion61,64. It has been demonstrated that hypoxia in combination with vascular endothelial growth factor (VEGF) and platelet-derived growth factor (PDGF) mediates the formation of the primary capillary plexus (angioblasts) as well as subsequent vasculogenesis (formation of new vessels) and angiogenesis (budding and sprouting from pre-existing vessels), leading to the establishment of a functional coronary artery tree44–47 (Figure 2).

**Box 1. Placental regulation of fetal oxygen tension**

A complex coordination exists between the fetal cardiovascular system and the placenta to ensure stable oxygen levels73,75–76. Low maternal blood oxygen levels, cardiovascular failure, or impairment of placenta formation and function can induce pathological fetal hypoxia88–90. In pathological hypoxia, the fetus responds by favoring circulation to vital organs such as the brain and heart. In addition, a concomitant activation-specific stress response (for example, Hif-dependent and vascular endothelial growth factor pathways80,81,83) occurs. Chronic fetal pathological hypoxia induces heart septation defects85, myocardial wall thinning, chamber dilation, and epicardium detachment86 as well as decrease of cardiomyocyte proliferation and increase of apoptosis81. Of note, in utero stress is also driven by other environmental stressors, including malnutrition that culminates with cardiac malformations and increased predisposition to adult chronic diseases80–85.

The murine heart is the first organ to function during development (E8.5)13,14, and the continuous myocardial contraction requires energy (adenosine triphosphate [ATP]) availability. About 80% of all energy produced by cardiac muscle is consumed by the mechanical activity of the heart, whereas heart morphogenesis relies on the remaining energy75. The developing heart relies primarily on carbohydrates (that is, glucose and lactate) as a source of ATP4,75. The placenta regulates the availability of metabolic substrates that are delivered to the embryonic/fetal circulation, whereas the fatty acids are blocked89. Glucose is transported to the cytosol of cardiomyocytes and used for glycolysis, glycogen synthesis, or pentosephosphate shunt (metabolic pathway parallel to glycolysis). Glycogen synthesis and storage in fetal cardiomyocytes have been shown to be important sources of phosphorylated glucose, which protects cardiomyocytes from hypoxia. During fetal development, cardiomyocytes have low mitochondrial content73,75. In summary, embryonic and fetal cardiac development occurs in a physiological hypoxic state, which is essential for the ability of cardiac progenitors to proliferate, self-renew, and differentiate (for example, immature cardiomyocyte hyperplasia or neo-angiogenesis).
Figure 2. Placenta-selected hypoxic and glycolytic microenvironment and its impact on the heart development and metabolic switch after birth. A color code and symbols were assigned and followed to define the microenvironment condition (depicted in the legend). E, embryonic day.
Early postnatal development represents a key transition in cardiac metabolism and complexity

The mammalian heart undergoes a marked increase in workload upon birth, and, in the mouse, the first few weeks of postnatal life are characterized by extensive ventricular remodeling coincident with switch from anaerobic metabolism (glycolysis) to aerobic metabolism (oxidative phosphorylation of fatty acids). Experiments performed in rabbits show that while circulating levels of fatty acid are high following birth, the switch from glycolysis to aerobic metabolism occurs only at the end of the first week, and the neonate heart retains an enriched ability to produce ATP by glycolytic metabolism (Figure 2). Postnatal myocardial growth involves an increase in hemodynamic demands and is characterized by a thickening and vascularization of the ventricular myocardial wall. Mouse postnatal growth follows three main steps: hyperplasia (postnatal day 0 to P4), the transitional phase when hyperplasia and hypertrophy processes occur simultaneously (P5 to P15), and hypertrophy (after P15). Studies in rodent models have shown that a last round of DNA synthesis and karyokinesis takes place without cytokinesis during the hypertrophy phase, culminating in the bi-nucleation of postnatal cardiomyocytes. In addition to cardiomyocyte maturation, there is a marked growth and maturation of the coronary vasculature. The perinatal period is characterized by angiogenesis and an expansion of the capillary network. This increase of the coronary tree is due to the proliferation of pre-existing capillary endothelial cells and increase of the capillary length as well as increases in the thickness, length, and branching of arterial and venous coronary vasculature.

Neonatal heart regeneration

In 2011, Porrello and colleagues demonstrated that the mouse neonatal heart regenerates in response to ventricular apex resection as well as experimentally induced myocardial infarction (MI). The regenerative process of the neonate heart is characterized by clot formation at the site of injury coupled with an inflammatory response followed by epicardial cell and cardiomyocyte proliferation, ultimately leading to a restoration of cardiac function. It is of interest that this regenerative capacity is lost during the first few days after birth, which, as noted above, corresponds to the time point during which the terminal differentiation of cardiomyocytes is concluded. Although these studies support the proposal that the mammalian neonatal heart possesses pronounced regenerative capacity, including the ability to replace cardiomyocytes, these results have been challenged by using the same experimental model in which limited cardiomyocyte proliferation and deficient neo-angiogenesis coupled with extensive scarring were observed. These different outcomes have been suggested to be due to technical variation, difficulties in tracking mouse cardiomyocyte proliferation during the first week of life, and timing of post-injury follow-up, where longer post-injury period allows more time for scar tissue deposition. Although there remains considerable debate regarding the efficiency of neonatal heart regeneration, it is generally agreed that the neonatal myocardium has some proliferative and angiogenic capacity which is lost in the adult heart. What might explain this more robust repair capacity during the first week of postnatal life in the mouse? As outlined in the previous section, the murine heart undergoes a metabolic switch to oxidative metabolism coupled with the maturation and bi-nucleation of cardiomyocytes, and the maturation of the coronary vasculature, which is driven by the availability of oxygen. This raises the hypothesis that low levels of oxygen during early postnatal life present a permissive context in which regeneration can occur.

Tissue repair in the adult heart: adult cardiac progenitor cells

The adult mammalian heart is one of the least-regenerative organs in the body; thus, injury, most notably MI, leads to a progressive decrease in heart function and ultimately results in heart failure. In the past two decades, several studies have shown that the uninjured adult heart replaces a low percentage of the total cardiomyocytes. Renewal of pre-existing cardiomyocytes is approximately 0.5% to 1% per year, a rate that declines with age. This low turnover rate implies that the vast majority of cardiomyocytes present at the conclusion of postnatal development remain throughout the entirety of adult life. It has been demonstrated that adult cardiomyocytes undergo a fourfold increase in turnover following injury, however, the origin of new cardiomyocytes remains unclear. Several studies have provided evidence for the existence of adult resident CPCs that are able to give rise to cardiomyocytes and non-myocytes that represent a potential source for heart regeneration. CPCs have been defined and isolated by the expression of different markers clustered in niches in specific regions such as the atria, apex, and epicardium (Table 1); however, it is still unclear whether these different subsets belong to the same cell population or represent different CPC populations or do both. Additionally, the developmental origin of adult CPCs remains largely unknown, except for some of the CPC subsets that maintain a protein signature highly related with the embryonic populations such as the PEO-derived and Isl1+ subsets (Table 1). After injury, CPCs are activated and give rise to different cell types (that is, myofibroblasts and smooth muscle cells and, to a lesser extent, endothelial cells and cardiomyocytes). Of note, the contribution of these cells to new cardiomyocytes during steady-state and injury is highly debated. One high-profile example is the...
c-kit expressing CPCs, which have been reported to possess proliferative capacity and the ability to differentiate into cardiomyocytes, whereas other studies have refuted these observations and observe little to no cardiomyogenic potential. Other studies have provided evidence demonstrating that the formation of new cardiomyocytes comes from pre-existing cardiomyocytes that proliferate in human steady-state young heart as well as in pathological conditions. Similarly, in the mouse, studies have suggested that new cardiomyocytes are derived from pre-existing cardiomyocytes that re-enter the cell cycle or arise from dedifferentiated myocytes. Regardless of whether one or both of these models are accurate, it is clear that the adult heart is unable to functionally restore the adult myocardium following cardiac injury. In addition to the inefficient renewal of cardiomyocytes after injury, little revascularization is observed in the adult heart following injury, which likely contributes to poor cardiac regeneration. Some studies revealed that endothelial cells, resident endothelial cells, or even fibroblasts can adopt an endothelial cell-like fate and contribute to angiogenesis in response to cardiac injury; however, the involvement of CPCs in this process remains largely controversial. In the next section, we consider whether the potential of the heart to neovascularize following injury is a key process for future therapeutic targeting for heart disease.

The role of the adult epicardium in cardiac repair

The adult myocardium is surrounded and protected by the epicardium, which consists of a single mesothelial cell layer. As described in the previous section, the fetal epicardium is a source of cellular components and paracrine signals that promote coronary vasculature formation and myocardial growth. Once the heart is mature, EPDCs progressively lose their capacity to undergo EMT and the epicardium enters a primarily quiescent state. This is of interest since Thymosin β-4 (Tβ-4) treatment results in re-expression of genes typically observed during fetal epicardial development in response to injury, including Wt1, Tbx18, and Raldh2, coupled with proliferation and EMT to form a thick layer of EPDC mesenchymal cells. EPDCs contribute to adventitial and interstitial fibroblasts and smooth muscle cells. (Figure 3), suggesting that the adult epicardium serves as a reservoir for mesenchymal progenitor cells.

It has been shown that thymosin β-4 (Tβ-4) treatment results in adult epicardial activation, migration, and differentiation of EPDCs into smooth muscle cells, endothelial cells, and fibroblasts. This is of interest since Tβ-4 is a key regulator of angiogenesis that acts through the stimulation of Vegf and stabilizing the hypoxia-inducible factor 1α (Hif-1α). It has been shown that Tβ-4 is expressed by the developing myocardium and regulates coronary vessel development (vasclogenesis, angiogenesis, and arteriogenesis) through activation of the epicardium. Conversely, Banerjee and colleagues could not confirm these results and claimed there is no link between Tβ-4 and angiogenesis during heart morphogenesis.

---

**Table 1. Different identified adult cardiac progenitor cell populations.**

| Adult cardiac progenitor cells | Embryonic origin | Progenies | References |
|-------------------------------|-----------------|----------|------------|
| Identification/isolation by cell surface markers | | | |
| c-kit+ | Unknown | Cardiomyocytes, endothelial cells, and smooth muscle cells | 146–149,150 |
| Sca-1+ | Unknown | Cardiomyocytes, endothelial cells, smooth muscle cells, and fibroblasts | 151–156 |
| Islet-1+ | SHF proposed | Smooth muscle cells, endothelial cells, parasympathetic neurons, sinoatrial node cells, and cardiomyocytes | 20,141–143,157,158 |
| Platelet-derived growth factor receptor alpha+ (PDGFRα+) | Unknown | Smooth muscle cells and endothelial cells | 159,160 |
| Mesangioblasts | Unknown | Cardiomyocytes | 161,162 |
| Epicardial-derived progenitor cells | PEO | Coronary smooth muscle cells and adventitial and interstitial fibroblasts | 56,138,140,163 |
| Identification by dye-efflux function | | | |
| Side population | Unknown | Cardiomyocytes, endothelial cells, smooth muscle cells, and fibroblasts | 151,164–167 |
| Isolation by in vitro assays | | | |
| Cardiospheres and cardiosphere-derived cells | Unknown | Cardiomyocytes | 168–170 |
| Cardiac-resident colony-forming units-fibroblasts (cCFU-Fs) | PEO proposed | Fibroblasts | 139 |
different transgenic models used in the two studies may explain the discrepancy of the results obtained. Tβ-4 is also able to promote the vascular potential of adult EPDCs, which contributes to an enhanced cardiomyocyte survival after injury.\textsuperscript{58,143,205,207} Irrespective of the angiogenic properties of Tβ-4 during development, additional studies emphasize the involvement of epicardial cells and EPDCs in vasculature formation after cardiac injury. Wagner and colleagues\textsuperscript{208} demonstrated that Wt1, the epicardial master transcription factor, is expressed in the coronary vessels of injured heart, suggesting a role for Wt1 in the vascular growth after cardiac injury. Additionally, proangiogenic factors, such as VEGF and FGF, are secreted by EPDCs during the cardiac repair process, promoting the growth and survival of the heart vasculature.\textsuperscript{195}

In addition to providing a protective role for the heart, the epicardium serves as a reservoir for CPCs that are activated in response to injury. As such, the adult epicardium merits future research for targeting and ameliorating mammalian heart repair.

**The adult cardiac cellular microenvironment**

The heart continually adapts to changing workloads brought about by aging, physical activities, and disease. The heart produces energy using multiple metabolic substrates such as...
fatty acids, glucose, ketone bodies, lactate, and amino acids. It is estimated that the heart uses between 3.5 and 5 kg of ATP every day. Fatty acids are more energy-dense and thus provide more ATP molecules per consumed carbon as compared with the other substrates; however, fatty acid pathway requires more oxygen. A healthy heart uses primarily long-chain fatty acids, which provide 60% to 70% of the produced ATP to power the muscle contraction, while the remaining energy is derived from carbohydrate metabolism. To meet this high ATP demand, the heart requires oxygen. When the body is at rest, myocardial oxygen consumption is greater than the oxygen consumption of any other organ of the body. Coronary circulation ensures the delivery of metabolic substrates and oxygen to the myocardium. Under physiological conditions, 90% of the ATP is produced through mitochondrial oxidative phosphorylation (fatty acids and glucose), which is an oxygen-dependent process. However, hypoxic conditions resulting from either physiological states (for example, during exercise) or pathological states (for example, coronary artery disease with mild ischemia) require the glycolytic pathway in order to produce ATP.

Although the adult heart is highly reliant on the availability of high levels of oxygen, the ventricles have a non-uniform distribution of oxygen across the wall with an oxygen tension gradient: lower tension in the extremities—that is, subendocardial (pO_2 = 10 mmHg) and subepicardial (pO_2 = 18 mmHg) regions—and higher tension in the middle of the myocardium (pO_2 = 38 mmHg). Reflecting the different levels of oxygen tension in the heart, Hif-1α, known to be stable under hypoxic conditions, is detected at high levels in the epicardium and subepicardium. Although the high levels of environmental oxygen and adult stem cell niches (for example, bone marrow) are characterized by low concentration of oxygen tension, this favors glycolytic metabolism. Although it has been shown to be essential, the exact mechanism of glycolytic metabolism and hypoxia in the maintenance of stemness properties is not well understood. A protective mechanism of the adult stem cells against reactive oxygen species (ROS) has been implicit, but more studies are needed. As described previously, the epicardium represents a reservoir of adult CPCs and, like other adult stem cell niches, displays a low oxygen tension and relies on glycolysis-dependent metabolic pathways (cytoplasmic glycolysis) (Figure 3).

Potential advantages of environmental manipulation to stimulate epicardial cells to a more efficient repair

Cardiovascular diseases are a major cause of human mortality. At present, long-term and effective treatments are missing. It is noteworthy that in high-altitude regions, cardiovascular diseases are less prevalent, which has been attributed to continuous low-level hypoxia. Hypoxia activates an evolutionarily conserved adaptive process that allows mammals to cope with restricted oxygen tension. Indeed, as already described in this review, the hypoxic environment promotes a metabolic switch from aerobic mitochondrial metabolism to anaerobic cytoplasmic glycolysis, which is an essential feature of heart development driving cell proliferation, self-renewal, and differentiation as well as of specific cardiac processes such as ventricular wall expansion through cardiomyocyte hyperplasia or stimulation of angiogenesis and formation of coronary vasculature. This feature of adult progenitor cells allows ATP production in the absence of oxygen, providing an advantage in a hypoxic environment. Additionally, it has become clearer that while oxidative metabolism is more efficient for producing ATP, cellular senescence and cell cycle arrest are a consequence of the resultant oxidative stress.

Although this review is focused on mammalian heart biology, it is important to note that lower vertebrates such as the zebrafish maintain a capacity to fully regenerate large domains of damaged myocardium through the proliferation of pre-existing cardiomyocytes, which is similarly triggered by a hypoxic protective response. These results prompted Sadek and colleagues to explore the response of the mammalian heart to injury under hypoxic environmental conditions. Myocardial infarcted mice exposed for two weeks to hypoxia displayed a reduction of myocardial fibrosis and improved ventricular function. Sadek and colleagues proposed that the gradual reduction of environmental oxygen promotes glycolytic metabolism and reduces oxidative phosphorylation. This metabolic switch decreases ROS and cellular senescence and promotes the activation of proliferation of a small fraction of pre-existing cardiomyocytes; however, it is also likely that additional progenitor populations contribute to this response. Additionally, Sadek and colleagues noted that there is a marked cardiac vascular network expansion (that is, the increase of coronary collaterals and capillary size) in the infarcted area after exposure to low levels of oxygen as compared with controls. As discussed above, the angiogenesis is dependent upon the oxygen tension and thus on the metabolic status of the microenvironment.

Emerging evidence suggests that the epicardium is an important source of stromal progenitor cells during development and that it maintains this capacity in the adult, potentially because of the particular hypoxic and glycolytic environment preserved in this adult heart compartment. While chronic exposure to high altitude carries collateral health risks in humans, it is tempting to propose that low oxygen tension can maintain the epicardium in an activated state, thus preserving perinatal plasticity, which in turn leads to improved cardiac repair. Further investigation of the adult CPCs and their microenvironment will provide important leads for improving cardiac repair following injury.

Taken altogether, while robust heart regeneration in the adult has yet to be achieved through therapeutic intervention, an understanding of the environmental clues and metabolic state of the developing heart will provide essential clues for novel therapeutic approaches. In the short term, it may be important to reconsider methodologies used during patient recovery following heart injury or insufficiency or both. Specifically, a more efficient angiogenic repair coupled with reduced scar tissue formation may result from promoting a more “embryonic-like” cardiac tissue environment during the repair process. How this can be achieved in a clinical setting and what additional
clues can be translated to the clinic will be an interesting focus for the next decade.

Authors’ contributions

AS and MV contributed to original draft preparation and to reviewing and editing. DS contributed to conceptualization and to reviewing and editing.

References

1. Nag AC: Study of non-muscle cells of the adult mammalian heart: a fine structural analysis and distribution. Cytobios. 1980; 28(10):41–61. PubMed Abstract
2. Baraneev I, Yekitska K, Borg TK, et al.: Dynamic interactions between myocytes, fibroblasts, and extracellular matrix. Ann N Y Acad Sci. 2006; 1080:76–84. PubMed Abstract | Publisher Full Text
3. Saga Y, Miyagawa-Tomita S, Takagaki A, et al.: MesP1 is expressed in the heart precursor cells and required for the formation of a single heart tube. Development. 1999; 126(15):3437–3447. PubMed Abstract
4. Kajtaz T, Nishii K, Ueoka H, et al.: Recent improvement in lung cancer screening: a comparison of the results carried out in two different time periods. Acta Med Okayama. 2006; 60(3):173–179. PubMed Abstract | Publisher Full Text
5. Jiang X, Rowitch DH, Soriano P, et al.: Fate of the mammalian cardiac neural crest. Development. 2000; 127(6):1607–1616. PubMed Abstract
6. Vrignon S, Challic CE: The origin of the epicardium and the embryonic myocardial circulation in the mouse. Anat Rec. 1981; 201(4):157–168. PubMed Abstract | Publisher Full Text
7. Pérez-Pomares JM, Phelps A, Sedmerova M, et al.: Epicardial-like cells on the distal arterial end of the cardiac outflow tract do not derive from the proepicardium but are derivatives of the cephalic pericardium. Dev Dyn. 2003; 227(1):56–68. PubMed Abstract | Publisher Full Text
8. Wu SM, Fujikawa Y, Cibulsky SM, et al.: Developmental origin of a bipotential myocardial and smooth muscle cell precursor in the mammalian heart. Cell. 2000; 127(6):1137–1150. PubMed Abstract | Publisher Full Text
9. Moihtac SM, Eimer M, Kelly RG, et al.: The clonal origin of myocardial cells in different regions of the embryonic mouse heart. Dev Cell. 2004; 6(5):685–698. PubMed Abstract | Publisher Full Text
10. Saga Y, Kloska S, Miyagawa-Tomita S: Mesp1 expression is the earliest sign of cardiovascular development. Trends Cardiovasc Med. 2000; 10(8):345–352. PubMed Abstract | Publisher Full Text
11. Harvey RP: Patterning the vertebrate heart. Nat Rev Genet. 2002; 3(7):544–556. PubMed Abstract | Publisher Full Text
12. Zaffran S, Kelly RG, Moihtac SM, et al.: Right ventricular myocardium derives from the anterior heart field. Circ Res. 2004; 95(3):261–268. PubMed Abstract | Publisher Full Text
13. Nishi K, Shibata Y: Mode and determination of the initial contraction stage in the mouse embryo heart. Anat Embryol (Berl). 2006; 211(2):95–100. PubMed Abstract | Publisher Full Text
14. Navaratnam V, Kaufman MH, Skepper JN, et al.: Differentiation of the myocardial rudiment of mouse embryo: an ultrastructural study including freeze-fracture replication. J Anat. 1980; 146:65–85. PubMed Abstract | Publisher Full Text
15. Kelly RG, Broen NA, Buckingham ME: The arterial pole of the mouse heart forms from Fgf10-expressing cells in pharyngeal mesoderm. Dev Cell. 2001; 1(3):435–440. PubMed Abstract | Publisher Full Text
16. Cai CL, Liang X, Shi Y, et al.: Isl1 identifies a cardiac progenitor population that proliferates prior to differentiation and contributes a majority of cells to the heart. Dev Cell. 2003; 5(6):877–889. PubMed Abstract | Publisher Full Text | Free Full Text | F1000 Recommendation
17. Galli D, Dorméjus JN, Zaffran S, et al.: Atrial myocardium derives from the posterior region of the second heart field, which acquires left-right identity as PitX2c is expressed. Development. 2008; 135(6):1157–1167. PubMed Abstract | Publisher Full Text
18. Verzi MP, McCulley DJ, De Val S, et al.: The right ventricle, outflow tract, and ventricular septum comprise a restricted expression domain within the secondary/anterior heart field. Dev Biol. 2005; 287(1):134–145. PubMed Abstract | Publisher Full Text
19. Buckingham M, Moihtac S, Zaffran S: Building the mammalian heart from two sources of myocardial cells. Nat Rev Genet. 2005; 6(11):826–835. PubMed Abstract | Publisher Full Text
20. Moretti A, Caron L, Nakano A, et al.: Multipotent embryonic Isl1+ progenitor cells lead to cardiac, smooth muscle, and endothelial cell diversification. Cell. 2006; 127(6):1115–1165. PubMed Abstract | Publisher Full Text | F1000 Recommendation
21. Sun Y, Liang X, Najafi N, et al.: Isl1 is expressed in distinct cardiovascular lineages, including pacemaker and coronary vascular cells. Dev Biol. 2007; 304(1):286–296. PubMed Abstract | Publisher Full Text | Free Full Text
22. Watanabe Y, Buckingham M: The formation of the embryonic mouse heart: heart fields and myocardial cell lineages. Ann N Y Acad Sci. 2010; 1188(1):15–24. PubMed Abstract | Publisher Full Text
23. Walsø KL, Hutson MR, Ward CC, et al.: Secondary heart field contributes myocardium and smooth muscle to the arterial pole of the developing heart. Dev Biol. 2005; 281(1):78–90. PubMed Abstract | Publisher Full Text | Free Full Text
24. Harmon AW, Nakano A: Nkx2-5 lineage tracing visualizes the distribution of second heart field-derived aortic smooth muscle. Genesis. 2013; 51(12):862–869. PubMed Abstract | Publisher Full Text | Free Full Text
25. Ma Q, Zhou B, Pu WT: reassessment of Isl1 and Nkx2-5 cardiac fate maps using a Gata4-based reporter of Cre activity. Dev Biol. 2008; 323(1):98–104. PubMed Abstract | Publisher Full Text | Free Full Text
26. Mindt CS: On a Hitherto Unrecognized Form of Blood Circulation without Capillaries in the Organs of Vertebrates. J. Boston Soc Med Sci. 1900; 4(6):133–134. PubMed Abstract | Free Full Text
27. Sedmera D, Piexidter T, Vuilemim M: Developmental patterning of the myocardium. Anat Rec. 2000; 258(4):319–337. PubMed Abstract | Publisher Full Text
28. Sedmera D, Devkova M, DeAmeleida A: Spatialtemporal pattern of commitment to slowed proliferation in the embryonic mouse heart indicates progressive developmental of the cardiac conduction system. Anat Rec A Discov Mol Cell Evol Biol. 2003; 274(1):773–777. PubMed Abstract | Publisher Full Text
29. Sedmera D, Thompson RP: Myocyte proliferation in the developing heart. Dev Dyn. 2011; 240(6):1322–1334. PubMed Abstract | Publisher Full Text | Free Full Text
30. de Boer BA, van den Berg G, de Boer PA, et al.: Growth of the developing mouse heart: an interactive qualitative and quantitative 3D atlas. Dev Biol. 2012; 368(2):203–213. PubMed Abstract | Publisher Full Text | Free Full Text
31. Pérez-Pomares JM, Phelps A, Sedmerova M, et al.: Experimental studies on the spatialtemporal expression of WT1 and RALDH2 in the embryonic avian heart: a model for the regulation of myocardial and valvuloseptal development by epicardially derived cells (EPCs). Dev Biol. 2002; 247(2):307–326. PubMed Abstract | Publisher Full Text
32. Chen T, Chang TC, Kang JO, et al.: Epicardial induction of fetal cardiomyocyte proliferation via a retinoic acid-inducible trophic factor. Dev Biol. 2002; 250(1):198–207. PubMed Abstract | Publisher Full Text
33. Pennisi DJ, Ballard VL, Mikawa T: Epicardium is required for the full rate of Page 11 of 17

Grant information

The laboratory is supported by the Laboratoire d’Excellence Revive (Investissement d’Avenir, ANR-10-LABX-73), the Fondation Leducq (grant 13CVD01; CardioStemNet project), and Agence Nationale pour la Recherche (ANR) grant RHU CARMMA (ANR-15-RHU-0003).

The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.
myocyte proliferation and levels of expression of myocyte mitogenic factors FGFR2 and its receptor, FGFR-1, but not for transmural myocardial patterning in the embryonic chick heart. Dev Dyn. 2003; 228(2): 161–172.

Baldwin HS: Early embryonic vascular development. Cardiovasc Res. 1996; 31 Spec No: 34–45.

Drake CJ, Fleming PA: Vasculogenesis in the day 6.5 to 9.5 mouse embryo. Blood. 2000; 95(6): 1671–1679.

Mispfel AM, Boyle SC, Tompkins KL, et al.: Endocardial cells are a distinct endothelial lineage derived from Fli1+ multipotent cardiovascular progenitors. Dev Biol. 2009; 332(1): 78–89.

Gassmann M, Casagrande F, Ortili D, et al.: Ablernant neural and cardiac development in mice lacking the ErbB4 neuregulin receptor. Nature. 1995; 378(6555): 390–394.

LeeKF, Simon H, Chen H, et al.: Requirement for neuregulin receptor erbB2 in neural and cardiac development. Nature. 1995; 378(6555): 384–388.

Wagner M, Siddiqui MA: Signal transduction in early heart development (II): ventricular chamber specification, trabeculation, and heart valve formation. Exp Biol Med (Maywood). 2007; 232(7): 866–880.

Stankunas K, Hang CT, Tsun ZY, et al.: ADAMTSL5 to maintain the microenvironment for myocardial morphogenesis. Dev Cell. 2008; 14(2): 298–311.

Rychterova V: Principle of growth in thickness of the heart ventricular wall in the chick embryo. Folia Morphol (Praha). 1971; 19(3): 263–272.

Seidema D, Poxieder T, Hu N, et al.: Developmental changes in the myocardial architecture of the chick. Anat Rec. 1997; 248(3): 421–432.

Wu B, Zhang Z, Liu W, et al.: Endocardial cells form the coronary arteries by angiogenesis through myocardial-endoocardial VE-cadherin signaling. Cell. 2012; 151(1): 1083–1096.

Tian X, Hu T, Zhang H, et al.: Vessel formation. De novo formation of a distinct coronary vascular population in neonatal heart. Science. 2014; 345(6192): 90–94.

Combs MD, Yuzen KE: Heart valve development: regulatory networks in development and disease. Circ Res. 2009; 105(9): 469–481.

Ranger AM, Grunsky MJ, Hodge MR, et al.: The transcription factor NF-ATc is essential for cardiac valve formation. Nature. 1998; 392(6672): 186–190.

Person AD, Klewer SE, Runyan RB: Cell biology of cardiac cushion development. Int Rev Cytol. 2003; 234: 287–335.

Smarr BS, Kern CB, Wessels A: Origin and fate of cardiac mesenchyme. Dev Dyn. 2008; 237(10): 2804–2819.

Eisenberg LM, Markwald RR: Molecular regulation of atrioventricular valvuloseptal morphogenesis. Circ Res. 1995; 77(1): 1–6.

de Lange FJ, Mooiman AF, Anderson RH, et al.: Lineage and morphogenetic analysis of the cardiac valves. Circ Res. 2004; 94(6): 645–654.

Wu B, Wang Y, Liu W, et al.: Mafcb1 coordinates valve endocardial cell lineage development and cardiac valve formation. Circ Res. 2011; 109(2): 183–192.

Kirby ML, Waldo KL: Neural crest and cardiovascular patterning. Circ Res. 1995; 77(2): 211–215.

Schulte I, Schlueter J, Abu-Issa R, et al.: Morphological and molecular left-right asymmetries in the development of the proepicardium: a comparative analysis on mouse and chick embryos. Dev Dyn. 2007; 236(4): 664–695.

van Wijk B, van den Berg G, Abu-Issa R, et al.: Epicardium and myocardium separate from a common precursor pool by crosstalk between bone morphogenetic protein- and fibroblast growth factor-signalizing pathways. Circ Res. 2009; 105(5): 431–441.

Wessels A, Pérez-Pomares JM: The epicardium and epicardially derived cells (EPDCs) as cardiac stem cells. Anat Rec A Discov Mol Cell Evol Biol. 2004; 278(1): 43–57.
to the myocardium of the chick embryo heart? A quail-chick chimera study tracing the fate of the epicardial primordium. Anat Rec. 1999; 255(2): 212–226.

PubMed Abstract

193. Muñoz-Chápuli R, Macias D, González-Iraite M, et al.: [The epicardium and epicardial-derived cells: multiple functions in cardiac development]. Rev Esp Cardiol. 2002; 55(10): 1070–1082.

PubMed Abstract

194. Manner J, Schlueter J, Brand T: Experimental analyses of the function of the proepicardium using a new microsurgical procedure to induce loss-of-proepicardial-function in chick embryos. Dev Dyn. 2005; 233(4): 1445–1463.

PubMed Abstract | Pubisher Full Text

195. Zhou B, Honor LB, He H, et al.: Adult mouse epicardium modulates myocardial injury by secreting paracrine factors. J Clin Invest. 2011; 121(5): 1894–1904.

PubMed Abstract | Pubisher Full Text | Free Full Text | F1000 Recommendation

196. von Gise A, Pu WT: Endocardial and epicardial epithelial to mesenchymal transitions in heart development and disease. Circ Res. 2012; 110(2): 1628–1645.

PubMed Abstract | Pubisher Full Text | Free Full Text | F1000 Recommendation

197. Riley PR: An epicardial floor plan for building and rebuilding the mammalian heart. Curr Top Dev Biol. 2012; 100: 233–251.

PubMed Abstract | Pubisher Full Text

198. Smits AM, Dronkers E, Goumans MJ: The epicardium as a source of multipotent adult cardiac progenitor cells:Their origin, role and fate. Pharmacol Res. 2010; 62: 127–135.

PubMed Abstract | Pubisher Full Text | Free Full Text | F1000 Recommendation

199. Gillenberger-de Groot AC, Winter EM, Poelmann RE: Epicardium-derived cells (EPDCs) in development, cardiac disease and repair of ischemia. J Mol Cell Cardiol. 2010; 49(5): 1056–1060.

PubMed Abstract | Pubisher Full Text | Free Full Text | F1000 Recommendation

200. Duan J, Ghenge C, Liu D, et al.: Wnt5a/catenin injury response activates the epicardium and cardiac fibroblasts to promote cardiac repair. EMBO J. 2012; 31(2): 429–442.

PubMed Abstract | Pubisher Full Text | Free Full Text | F1000 Recommendation

201. Brotsch CM, Kanisicak O, van Berlo JH: Differential expression of embryonic epicardial progenitor markers and localization of cardiac fibrosis in adult ischemic and hypertensive heart disease. J Mol Cell Cardiol. 2013; 65: 108–119.

PubMed Abstract | Pubisher Full Text | Free Full Text

202. Limana F, Zacheo A, Mocini D, et al.: Identification of myocardial and vascular precursor cells in human and mouse epicardium. Circ Res. 2007; 101(12): 1255–1265.

PubMed Abstract | Pubisher Full Text | Free Full Text

203. Bollini S, Vieira JM, Howard S, et al.: Re-activated adult epicardial progenitor cells are a heterogeneous population molecularly distinct from their embryonic counterparts. Stem Cells Dev. 2014; 23(15): 1719–1739.

PubMed Abstract | Pubisher Full Text | Free Full Text | F1000 Recommendation

204. Jo JO, Kim SR, Bae MK, et al.: Thymosin β4 induces the expression of vascular endothelial growth factor (VEGF) in a hypoxia-inducible factor (HIF)-1-dependent manner. Biochem Biophys Acta. 2013; 1831(11): 1244–1251.

PubMed Abstract | Pubisher Full Text | Free Full Text | F1000 Recommendation

205. Banerjee I, Moore Morris T, Evans SM, et al.: Thymosin β4 is not required for embryonic viability or vascular development. Circ Res. 2013; 112(3): e25–28.

PubMed Abstract | Pubisher Full Text | Free Full Text | F1000 Recommendation

206. Marks ED, Kumar A: Thymosin β4: Roles in Development, Repair, and Engineering of the Cardiovascular System. Vitam Horm. 2016; 102: 227–249.

PubMed Abstract | Pubisher Full Text | Free Full Text | F1000 Recommendation

207. Wagner KD, Wagner N, Bondke A, et al.: The Wilms’ tumor suppressor WT1 is expressed in the coronary vasculature after myocardial infarction. PASEJ B. 2002; 16(9): 1117–1119.

PubMed Abstract | Pubisher Full Text

208. Bing RJ, Siegel A, Unger I, et al.: Metabolism of the human heart. II. Studies on fat, ketone and amino acid metabolism. Am J Med. 1954; 18(4): 504–515.

PubMed Abstract | Pubisher Full Text

209. Opie LH: Heart Physiology: From Cell to Circulation. (Lippincott Williams & Wilkins, Philadelphia, USA ed. 4th Edition, 2004.

Reference Source

210. Opie LH: The metabolic vicious cycle in heart failure. Lancet. 2004; 364(9447): 1733–1734.

PubMed Abstract | Pubisher Full Text

211. Taegtmeyer H, Wilson CR, Razaighi P, et al.: Metabolic energetics and genetics in the heart. Ann N Y Acad Sci. 2005; 1047: 209–218.

PubMed Abstract | Pubisher Full Text

212. Heathcote HC, Clarke K: Metabolism, hypoxia and the diabetic heart. J Mol Cell Cardiol. 2011; 50(4): 598–605.

PubMed Abstract | Pubisher Full Text

Page 15 of 17
214. van der Vusse GJ, Glatz JF, Stam HC, et al.: Fatty acid homeostasis in the normoxic and ischemic heart. Physiol Rev. 1992; 72(4): 881–940. PubMed Abstract | Publisher Full Text

215. Giordano FJ: Oxygen, oxidative stress, hypoxia, and heart failure. J Clin Invest. 2005; 115(3): 500–508. PubMed Abstract | Publisher Full Text

216. Neely JR, Denton RM, Englund PJ, et al.: The effects of increased heart work on the tricarboxylate cycle and its interactions with glycolysis in the perfused rat heart. Biochem J 1972; 126(1): 147–159. PubMed Abstract | Publisher Full Text

217. Taegtmeyer H, Golman L, Sharma S, et al.: Linking gene expression to function: metabolic flexibility in the normal and diseased heart. Ann N Y Acad Sci. 2004; 1015: 202–213. PubMed Abstract | Publisher Full Text

218. Moss AJ: Intramyocardial oxygen tension. Circ Res. 1968; 2(3): 314–318. PubMed Abstract | Publisher Full Text

219. Kocabas F, Mahmoud AI, Sosic D, et al.: The hypoxic epicardial and subepicardial microenvironment. J Cardiovasc Transl Res. 2012; 5(5): 654–665. PubMed Abstract | Publisher Full Text | F1000 Recommendation

220. Semenza GL: Hypoxia-inducible factor 1 and cardiovascular disease. Annu Rev Physiol. 2014; 76: 39–56. PubMed Abstract | Publisher Full Text

221. Chow DC, Wenning LA, Miller WM, et al.: Modeling PO2 distributions in the bone marrow hematopoietic compartment. II. Modified Kroghian models. Biophys J. 2001; 81(2): 685–696. PubMed Abstract | Publisher Full Text | Free Full Text

222. Parmar K, Mauch P, Vergilio JA, et al.: Distribution of hematopoietic stem cells in the bone marrow according to regional hypoxia. Proc Natl Acad Sci U S A. 2007; 104(13): 5431–5436. PubMed Abstract | Publisher Full Text | Free Full Text

223. Simsek T, Kocabas F, Zhang J, et al.: The distinct metabolic profile of hematopoietic stem cells reflects their location in a hypoxic niche. Cell Stem Cell. 2010; 7(3): 580–590. PubMed Abstract | Publisher Full Text | Free Full Text

224. Urbanek K, Cesselli D, Rota M, et al.: Stem cell niches in the adult mouse heart. Proc Natl Acad Sci U S A. 2006; 103(24): 9226–9231. PubMed Abstract | Publisher Full Text | Free Full Text

225. Morrison SJ, Spradling AC: Stem cells and niches: mechanisms that promote stem cell maintenance throughout life. Cell. 2008; 132(4): 598–611. PubMed Abstract | Publisher Full Text | Free Full Text

226. Walker MR, Patel KK, Stappenbeck TS: The stem cell niche. J Pathol. 2009; 217(2): 169–180. PubMed Abstract | Publisher Full Text

227. Benjamin EJ, Blaha MJ, Chiuve SE, et al.: Heart Disease and Stroke Statistics-2017 Update: A Report From the American Heart Association. Circulation. 2017; 135(10): e146–e603. PubMed Abstract | Publisher Full Text | Free Full Text | F1000 Recommendation

228. Asemu G, Papoosek F, Ostadal B, et al.: Adaptation to high altitude hypoxia protects the rat heart against ischemia-induced arrhythmias. Involvement of mitochondrial KATP channel. J Mol Cell Cardiol. 1999; 31(10): 1821–1831. PubMed Abstract | Publisher Full Text | F1000 Recommendation

229. Faeh D, Gutzwiller F, Bopp M, et al.: Lower mortality from coronary heart disease and stroke at higher altitudes in Switzerland. Circulation. 2009; 120(6): 495–501. PubMed Abstract | Publisher Full Text

230. Bartsch P: Effects of living at higher altitudes on mortality: a narrative review. Aging Dis. 2014; 5(4): 274–280. PubMed Abstract | Publisher Full Text

231. Hoppeler H, Vogt M, Weibel ER, et al.: Response of skeletal muscle mitochondria to hypoxia. Exp Physiol. 2003; 88(1): 109–119. PubMed Abstract | Publisher Full Text

232. Gilbert-Kawai E, Coppel J, Court J, et al.: Sublingual microcirculatory blood flow and vessel density in Sherpas at high altitude. J Appl Physiol (1985). 2017; 122(4): 1011–1018. PubMed Abstract | Publisher Full Text | Free Full Text | F1000 Recommendation

233. Pass KD, Wilson LG, Keating MT: Heart regeneration in zebrafish. Science. 2006; 310(5746): 2188–2190. PubMed Abstract | Publisher Full Text | Free Full Text | F1000 Recommendation

234. Jopling C, Stemp E, Raya M, et al.: Zebrafish heart regeneration occurs by cardiomyocyte dedifferentiation and proliferation. Nature. 2010; 464(7288): 606–609. PubMed Abstract | Publisher Full Text | Free Full Text | F1000 Recommendation

235. Jopling C, Suárez G, Fauchere A, et al.: Hypoxia induces myocardial regeneration in zebrafish. Circulation. 2012; 126(25): 3017–3027. PubMed Abstract | Publisher Full Text

236. Nakada Y, Canseco DC, Thet S, et al.: Hypoxia induces heart regeneration in adult mice. Nature. 2017; 541(7636): 222–227. PubMed Abstract | Publisher Full Text | F1000 Recommendation

237. Kimura W, Nakada Y, Sadek HA: Hypoxia-induced myocardial regeneration. J Appl Physiol (1985). 2017; 123(6): 1676–1681. PubMed Abstract | Publisher Full Text | Free Full Text
Open Peer Review

Current Peer Review Status: ✔️ ✔️

Editorial Note on the Review Process

Faculty Reviews are review articles written by the prestigious Members of Faculty Opinions. The articles are commissioned and peer reviewed before publication to ensure that the final, published version is comprehensive and accessible. The reviewers who approved the final version are listed with their names and affiliations.

The reviewers who approved this article are:

1. Robert G. Kelly
   Developmental Biology Institute of Marseilles, Campus de Luminy Case 907, University of Aix-Marseille, CNRS UMR 7288, Marseilles Cedex 9, 13288, France
   Competing Interests: No competing interests were disclosed.

2. Jop H. van Berlo
   University of Minnesota, Lillehei Heart Institute and Stem Cell Institute, Minneapolis, MN, USA
   Competing Interests: No competing interests were disclosed.

The benefits of publishing with F1000Research:

- Your article is published within days, with no editorial bias
- You can publish traditional articles, null/negative results, case reports, data notes and more
- The peer review process is transparent and collaborative
- Your article is indexed in PubMed after passing peer review
- Dedicated customer support at every stage

For pre-submission enquiries, contact research@f1000.com