Cardiac Development

3

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Abstract

The first heart field (FHF), second heart field (SHF), cardiac neural crest (CNC), and proepicardial organ (PEO) are the four major embryonic regions involved in vertebrate heart development. They each make an important contribution to overall cardiac development with complex developmental timing and regulation. This chapter describes how these regions interact to form the final structure of the heart in relationship to the developmental timeline of human embryology.

Keywords

Human heart embryology • First heart field • Second heart field • Cardiac neural crest • Proepicardial organ • Cardiac development

3.1 Introduction to Human Heart Embryology and Development

The first heart field (FHF), second heart field (SHF), cardiac neural crest (CNC), and the proepicardial organ (PEO) are the four major embryonic regions involved in the process of vertebrate heart development (Fig. 3.1). They each make an important contribution to cardiac development with their own complex developmental timing and regulation (Table 3.1) [1, 2]. The heart is the first internal organ to form and function during vertebrate development, and many of the mechanisms of heart formation are molecularly and developmentally conserved [3–6]. The description presented here is based on development research from the chick, mouse, frog, and human model systems. Research conducted in the last decade has redefined the FHF which gives rise to the left ventricle and parts of the atria; furthermore, it has led to the exciting discovery of the SHF which gives rise to the outflow tract, right ventricle, and parts of the atria of the mature heart [7–18]. These discoveries were critical steps in helping us understand how the outflow tract of the heart forms, a cardiac structure where many congenital heart defects arise, and thus has important implications for the understanding and prevention of congenital heart disease [6, 15–19]. Great strides have also been made in understanding the contributions of both the CNC [20] and the PEO [15, 21, 22] to overall heart development.

3.2 First Heart Field Contribution to the Linear Heart Tube, Left Ventricle, and Atria

The cells that will become the heart are among the first cell lineages formed in the vertebrate embryo [23, 24]. By day 15 of human development, the primitive streak has formed [1] and the first mesodermal cells to migrate (gastrulate) through the primitive streak are also the cells fated to become myocytes or heart cells [25, 26] (Fig. 3.2). These mesodermal cells dedicated for heart development migrate to an anterior and lateral position where they initially form a bilateral FHF and a more medially located SHF [10, 11, 15, 16, 27]

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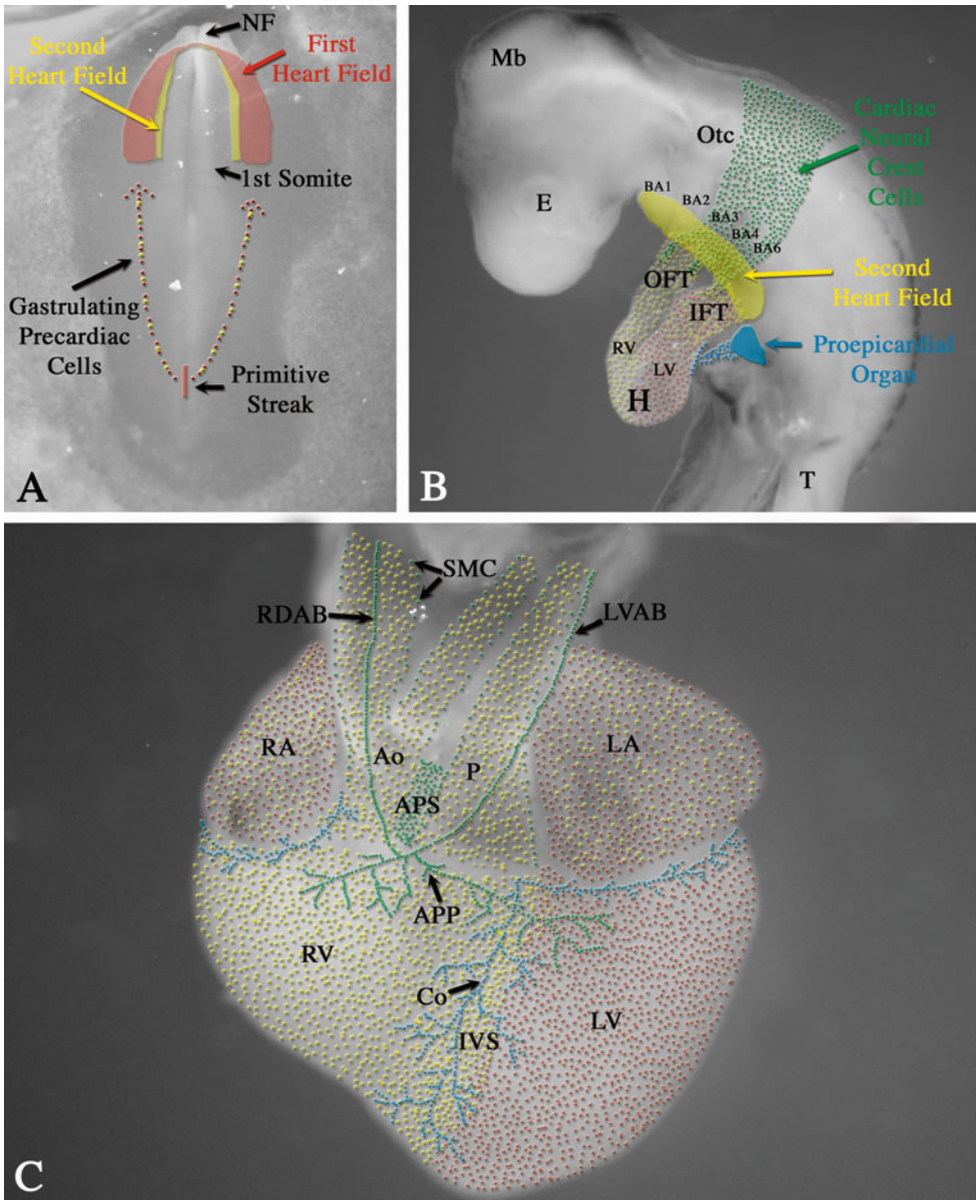

Fig. 3.1 The four major contributors to heart development illustrated in the chick model system: first heart field, second heart field, cardiac neural crest, and the proepicardial organ. (**A**) Day 1 chick embryo (equivalent to day 20 of human development). *Red* denotes first heart field cells and *yellow* denotes second heart field cells. (**B**) Day 2.5 chick embryo (equivalent to approximately 5 weeks of human development). Color code: *green* = cardiac neural crest cells; *red* = first heart field cells; *yellow* = second heart field cells; *blue* = proepicardial cells. (**C**) Day 8 chick heart (equivalent to approximately 9 weeks of human development). Color code: *green* = derivatives of the cardiac neural crest; *yellow* = derivatives of the second heart field; *red* = derivatives of the first heart field; *blue* = derivatives of the proepicardial organ. *Ao* aorta, *APP* anterior parasympathetic plexus, *APS* aorticopulmonary septum, *BA* branchial arch, *Co* coronary vessels, *E* eye, *H* heart, *IFT* inflow tract, *IVS* interventricular septum, *LA* left atrium, *LV* left ventricle, *LVAB* left ventral arterial branch of the Xth (vagal) cranial nerve, *Mb* midbrain, *NF* neural folds, *OFT* outflow tract, *Otc* otic placode, *P* pulmonary artery, *RA* right atrium, *RDAB* right dorsal arterial branch of the Xth (vagal) cranial nerve, *RV* right ventricle, *SMC* smooth muscles cells, *T* trunk

(Fig. 3.1A). Specifically, the posterior border of the bilateral FHF reaches down to the first somite in the lateral mesoderm on both sides of the midline [8, 28] (Fig. 3.1A). At day 18 of human development, the lateral plate mesoderm is split into two layers—somatopleuric and splanchnopleuric [1]. It is the splanchnopleuric mesoderm layer that contains the myocardial, smooth muscle, and endocardial cardiogenic precursors in the region of the FHF and SHF, as defined above. Presumptive endocardial cells delaminate from the splanchnopleuric mesoderm in the FHF and coalesce via vasculogenesis to form two lateral endocardial tubes [29]. During the third week of human development, two bilateral layers of

Table 3.1 Developmental timeline of human heart embryology

Human development (days)	Developmental process
0	Fertilization
1–4	Cleavage and movement down the oviduct to the uterus
5–12	Implantation of the embryo into the uterus
13–14	Primitive streak formation (midstreak level contains precardiac cells)
15–17	Formation of the three primary germ layers (gastrulation): ectoderm, mesoderm, and endoderm; midlevel primitive streak cells that migrate to an anterior and lateral position form the bilateral **first heart field** and a more medially located the **second heart field**
17–18	Lateral plate mesoderm splits into the somatopleuric mesoderm and splanchnopleuric mesoderm; splanchnopleuric mesoderm contains the myocardial and endocardial cardiogenic precursors in the region of the **first heart field** and **second heart field**
18–26	Neurulation (formation of the neural tube)
20	Cephalocaudal and lateral folding brings the bilateral endocardial tubes into the ventral midline of the embryo
21–22	Heart tube fusion
22	Heart tube begins to beat
22–28	Heart looping and the accretion of cells from the first and **second heart fields; proepicardial cells** invest the outer layer of the heart tube and eventually form the epicardium and coronary vasculature; neural crest migration starts
32–37	**Cardiac neural crest** migrates through the aortic arches and enters the outflow tract of the heart
57+	Outflow tract and ventricular septation complete
Birth	Functional septation of the atrial chambers, as well as the pulmonary and systemic circulatory systems

Most of the human developmental timing information is from *Larsen's Human Embryology* [1], except for the human staging of the second heart field and proepicardium which was correlated from other model systems [7–9, 30]

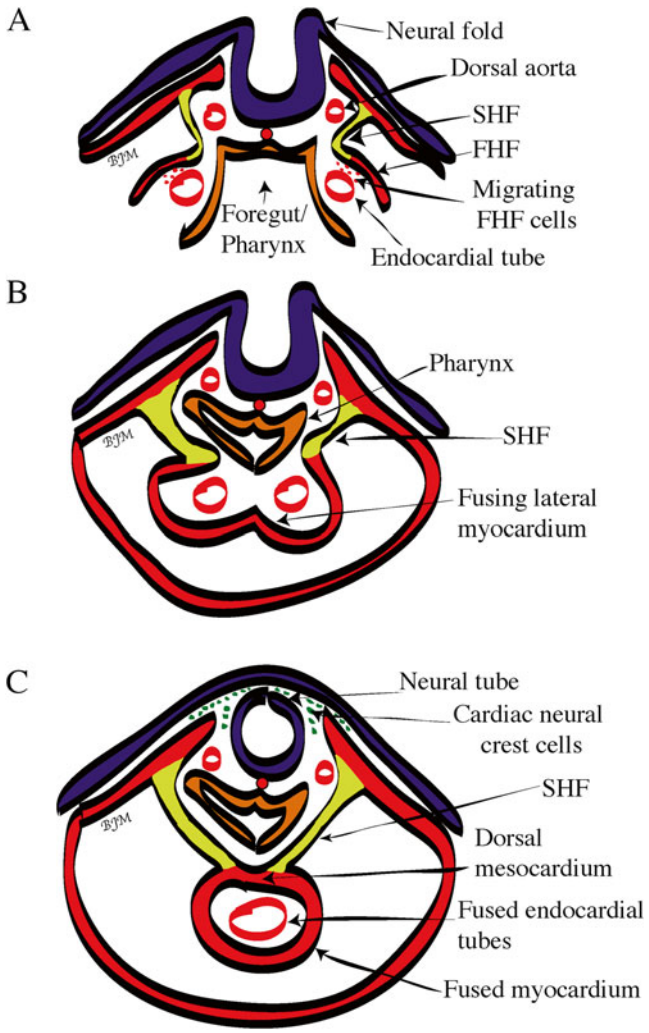

Fig. 3.2 Cross-sectional view of human heart tube fusion. (**A**) Day 20, cephalocaudal and lateral folding brings bilateral endocardial tubes into the ventral midline of the embryo. (**B**) Day 21, start of heart tube fusion. (**C**) Day 22, complete fusion, resulting in the beating primitive heart tube. Color code of the embryonic primary germ layer origin: *blue/purple* = ectoderm; *red* = mesoderm; *orange* = endoderm; *yellow* = second heart field. *FHF* first heart field, *SHF* second heart field

myocardium surrounding the endocardial tubes are brought into the ventral midline during closure of the ventral foregut via cephalic and lateral folding of the embryo [1] (Fig. 3.2A). The lateral borders of the myocardial mesoderm layers are the first heart structures to fuse, followed by the fusion of the two endocardial tubes which then form one endocardial tube surrounded by splanchnopleuric-derived myocardium (Fig. 3.2B, C). The medial borders of the myocardial mesoderm layers are the last to fuse [30]. Thus, the early heart is continuous with splanchnopleuric mesoderm across the dorsal mesocardium (Fig. 3.2C). This will eventually partially break down to form the ventral aspect of the linear heart tube with a posterior inflow (venous pole) and anterior outflow (arterial pole), as well as the dorsal wall of the pericardial cavity [18, 30]. During the fusion of the endocardial tubes, the myocardium secretes an extracellular (acellular) matrix (enriched in chondroitin sulfate, versican, heparan sulfate, hyaluronic acid, hyaluronan, and proteoglycans), forming the cardiac jelly layer separating the myocardium and endocardium [31]. By day 22 of human development, the linear heart tube begins to beat. As the human heart begins to fold and loop from day 22 to day 28 (described below), epicardial cells from the PEO will invest the outer layer of the heart tube (Figs. 3.1B and 3.3A), resulting in a heart tube with four primary layers: endocardium, cardiac jelly, myocardium, and epicardium [1] (Fig. 3.3B).

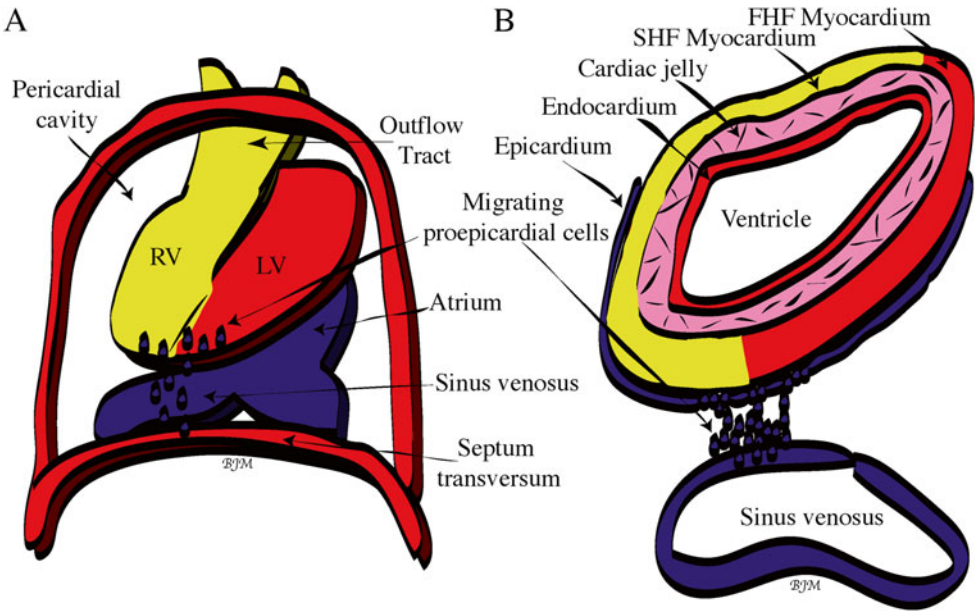

Fig. 3.3 Origin and migration of proepicardial cells. (**A**) Whole mount view of the looping human heart within the pericardial cavity at day 28. Proepicardial cells (*blue dots*) emigrate from the sinus venosus and possibly the septum transversum and then migrate out over the outer surface of the ventricles, eventually surrounding the entire heart. (**B**) Cross-sectional view of the looping heart showing the four layers of the heart: epicardium, myocardium, cardiac jelly, and endocardium. Color code: *yellow* = second heart field (SHF)-derived cells; *red* (within heart) = first heart field (FHF)-derived cells. *LV* left ventricle, *RV* right ventricle

3.3 Second Heart Field Contribution to the Outflow Tract, Right Ventricle, and Atria

A cascade of signals identifying the left and right sides of the embryo is thought to initiate the process of primary linear heart tube looping [32]. The primary heart tube loops to the right of the embryo and bends to allow convergence of the inflow (venous) and outflow (arterial) ends between day 22 and day 28 of human development (Fig. 3.4). This process occurs prior to the division of the heart tube into four chambers and is required for proper alignment and septation of the mature cardiac chambers. During the looping process, the primary heart tube increases dramatically in length (by four- to fivefold) on both the outflow and inflow poles via the addition of progenitor cells originating from the SHF (pharyngeal mesoderm) [7–18]. These multipotent progenitor cells within the developing heart give rise to myocardium, smooth muscle, and endothelial cells [12]. Previous experiments in the 1970s already revealed that the distal right ventricle and outflow tract (OFT) are added later to the looping heart by addition of cells lying outside the early heart [12, 33]. Researchers at that time, however, still assumed that the primary linear heart tube already contained all the cell lineages to build the adult heart. It was not until the rediscovery of these progenitor cells in 2001 (at the time termed *anterior heart field* or *secondary heart field*) that the clinical relevance of congenital heart defects was correlated to cells in this heart field—a big step in truly understanding heart development [7–9, 12]. The terms *anterior heart field* and *secondary heart field* are now considered to be a subpopulation of the SHF, a larger field of progenitor cells in pharyngeal mesoderm [12, 34]. The SHF is then contained within a larger field of multipotent cranial mesoderm (cardiocraniofacial field) that plays a critical role in development of both the arterial pole of the heart and craniofacial morphogenesis [12]. Specifically, the SHF (Figs. 3.1b and 3.2c) is located along the splanchnopleuric mesoderm (beneath the floor of the foregut) at the attachment site of the dorsal mesocardium [7–18]. During looping, the anterior SHF (previously termed *anterior heart field* or *secondary heart field*) cells undergo epithelial-to-myocardial transformation at the outflow (arterial) pole and add additional myocardial cells onto the then developing outflow tract, creating the great vessels (aorta and pulmonary trunk) and the right ventricle. This lengthening of the primary heart tube appears to be an important process for the proper alignment of the inflow and outflow tracts prior to septation. If this process does not occur normally, ventricular septal defects and malpositioning of the aorta may occur [30]. Recent evidence also indicates that the posterior SHF contributes to the inflow tract, creating parts of the left and right atria. Thus, the SHF contains two primary regions: (1) an anterior region or compartment that contributes to the outflow tract and (2) a posterior region or compartment that contributes to the inflow tract, as well as possibly the PEO [10, 15, 17, 35–37]. Defects in posterior SHF development result in conotruncal, atrial, and atrioventricular septal defects, major forms of congenital heart defects in humans [12].

By day 28 of human development, the chambers of the heart are in position and are demarcated by visible constrictions and expansions which denote the sinus venosus, common atrial chamber, atrioventricular sulcus, ventricular chamber, and conotruncus (proximal and distal outflow tract) [1, 30] (Fig. 3.4).

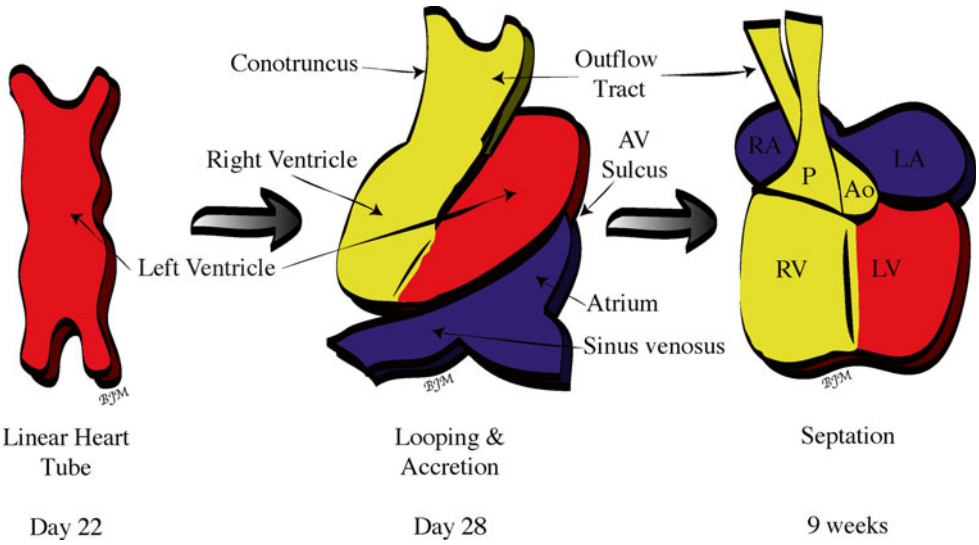

Fig. 3.4 Looping, accretion, and septation of the human primary linear heart tube. *Blue* (first heart field- and second heart field-derived cells) and *yellow* (second heart field-derived cells) regions represent tissue added during looping; *red* = first heart field-derived cells. *Ao* aorta, *AV* atrioventricular, *LA* left atrium, *LV* left ventricle, *P* pulmonary trunk, *RA* right atrium, *RV* right ventricle

3.4 Cardiac Neural Crest Contribution and Septation of the Outflow Tract and Ventricles

Once the chambers are in the correct position after looping, extensive remodeling of the primitive vasculature and septation of the heart can occur. The CNC is an extracardiac population of cells (from outside of the first or SHFs) that arise from the neural tube in the region of the first three somites up to the midotic placode level (rhombomeres 6, 7, and 8) (Fig. 3.5) [2, 38, 39]. CNC cells leave the neural tube during weeks 3–4 of human development and then migrate through aortic arches 3, 4, and 6 (Fig. 3.1b) and eventually into the developing outflow tract of the heart (during weeks 5–6). These cells are necessary for complete septation of the outflow tract and ventricles (completed by week 8 of human development), as well as the formation of the anterior parasympathetic plexus which contributes to cardiac innervation and regulation of heart rate [1, 2, 20, 38–42]. Recent evidence shows that CNC cells migrate to the venous pole of the heart as well and that their role is in the development of the parasympathetic innervation, the leaflets of the atrioventricular valves, and possibly the cardiac conduction system [43–45]. The primitive vasculature of the heart is bilaterally symmetrical but, during weeks 4–8 of human development, there is remodeling of the inflow end of the heart so that all systemic blood flows into the future right atrium [1]. In addition, there is also extensive remodeling of the initially bilaterally symmetrical aortic arch arteries into the great arteries (septation of the aortic and pulmonary vessels) that is dependent on the presence of the CNC [30, 46]. The distal outflow tract (truncus) septates into the aorta and pulmonary trunk via the fusion of two streams or prongs of CNC that migrate into the distal outflow tract. In contrast, the proximal outflow

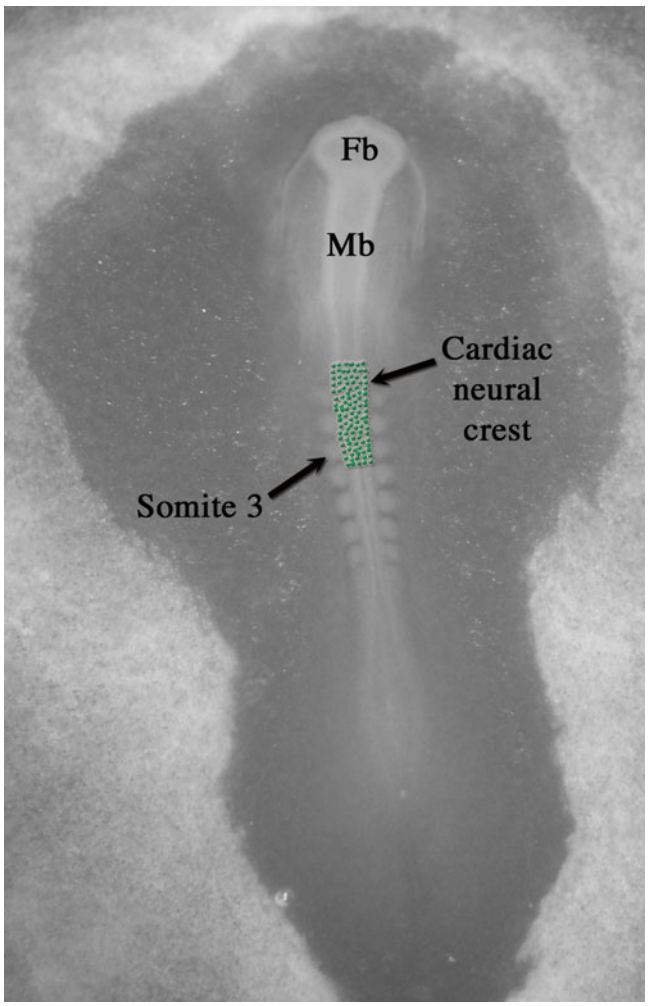

Fig. 3.5 Origin of the cardiac neural crest within a 34-h chick embryo. *Green dots* represent cardiac neural crest cells in the neural folds of hindbrain rhombomeres 6, 7, and 8 (the region of the first three somites up to the midotic placode level). *Fb* forebrain, *Mb* midbrain

tract septates by fusion of the endocardial cushions and eventually joins proximally with the atrioventricular endocardial cushion tissue and the ventricular septum [47, 48]. The endocardial cushions are formed by both atrioventricular canal and outflow tract endocardial cells that migrate into the cardiac jelly, forming bulges or cushions.

Despite its clinical importance, to this date, almost nothing is known about the molecular pathways that determine cell lineages in the CNC or regulate outflow tract septation [30, 49, 50]. However, it is known that if the CNC is removed before it begins to migrate, conotruncal septa completely fail to develop, and blood leaves both the ventricles through what is termed a *persistent truncus arteriosus*, a rare congenital heart anomaly that can be seen in humans [20, 40]. Failure of outflow tract septation may also be responsible for other forms of congenital heart disease including transposition of the great vessels, high ventricular septal defects, and tetralogy of Fallot [1, 20, 38, 40]. Additional information on these congenital defects can be found in Chap. 10.

The septation of the outflow tract (conotruncus) is tightly coordinated with the septation of the ventricles and atria to produce a functional heart [1, 51, 52]. All of these septa eventually fuse with the atrioventricular (AV) cushions that also divide the left and right AV canals and serve as a source of cells for the AV valves. Prior to septation, the right atrioventricular canal and right ventricle expand to the right, causing a realignment of the atria and ventricles so that they are directly over each other. This allows venous blood entering from the sinus venosus to flow directly from the right atrium to the presumptive right ventricle without flowing through the presumptive left atrium and ventricle [1, 30]. The new alignment also simultaneously provides the left ventricle with a direct outflow path to the truncus arteriosus and subsequently to the aorta.

Between weeks 4 and 7 of human development, the left and right atria undergo extensive remodeling and are eventually septated. Yet, during the septation process, a right-to-left shunting of oxygenated blood (oxygenated by the placenta) is created via a series of shunts, ducts, and foramens (Fig. 3.6). Prior to birth, the use of the pulmonary system is not necessary, but eventually a complete separation of the systemic and pulmonary circulatory systems will be required for normal cardiac and systemic function [1]. Initially, the right sinus horn is incorporated into the right posterior wall of the primitive atrium, and the trunk of the pulmonary venous system is incorporated into the posterior wall of the left atrium via a process called *intussusception*. At day 26 of human development, a crescent-shaped wedge of tissue called the septum primum begins to extend into the atrium from the mesenchyme of the dorsal mesocardium. As it grows, the septum primum diminishes the ostium primum, a foramen allowing the shunting of blood from the right to left atrium. However, programmed cell death near the superior edge of the septum primum creates a new foramen, the ostium secundum, which continues the right-to-left shunting

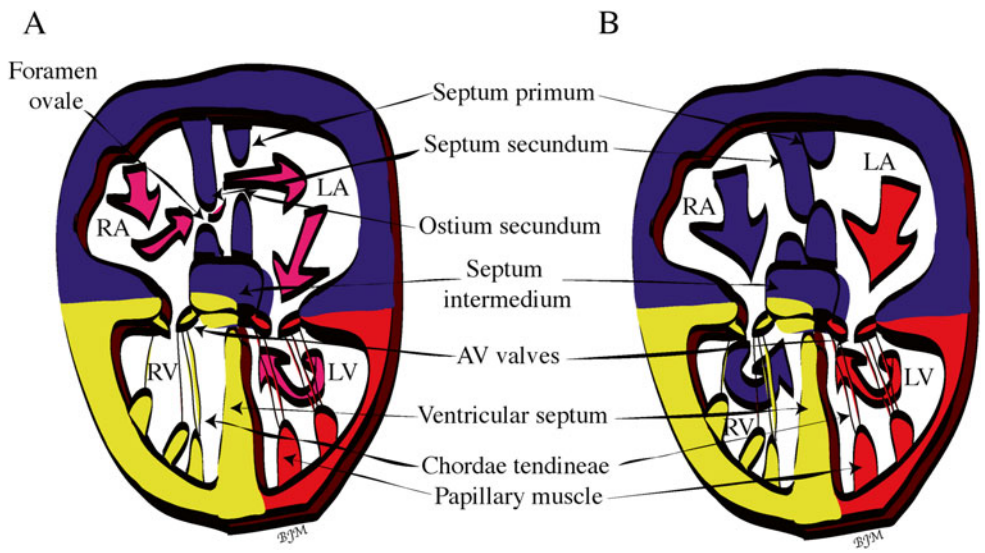

Fig. 3.6 Transition from fetal dependence on the placenta for oxygenated blood to self-oxygenation via the lungs. (**A**) Circulation in the fetal heart before birth. *Pink arrows* show right-to-left shunting of placentally oxygenated blood through the foramen ovale and ostium secundum. (**B**) Circulation in the infant heart after birth. The first breath of the infant and cessation of blood flow from the placenta cause final septation of the heart chambers (closure of the foramen ovale and ostium secundum) and thus separation of the pulmonary and systemic circulatory systems. *Blue arrows* show the pulmonary circulation and *red arrows* show the systemic circulation within the heart. Color code: *Blue* (first heart field- and second heart field-derived cells), *red* (first heart field-derived cells), and *yellow* (second heart field-derived cells). *AV* atrioventricular, *LA* left atrium, *LV* left ventricle, *RA* right atrium, *RV* right ventricle

of oxygenated blood. An incomplete, ridged septum secundum with a foramen ovale near the floor of the right atrium forms next to the septum primum, both of which fuse with the septum intermedium of the AV cushions [1]. At the same time as atrial septation is beginning, about the end of the fourth week of human development, the muscular ventricular septum begins to grow toward the septum intermedium (created by the fusion of the atrioventricular cushions), creating a partial ventricular septum. By the end of the ninth week of human development, the outflow tract septum has grown down onto the upper ridge of this muscular ventricular septum and onto the inferior endocardial cushion, completely separating the right and left ventricular chambers.

It is not until after birth, however, that the heart is functionally septated within the atrial region. At birth, dramatic changes in the circulatory system occur due to the transition from fetal dependence on the placenta for oxygenated blood to self-oxygenation via the lungs. More specifically, during fetal life, only small amounts of blood (about 5 % of the cardiac output) are flowing through the pulmonary system because the fluid-filled lungs create high flow resistance, resulting in low-volume flow into the left atrium from the pulmonary veins. This allows the high-volume blood flow coming from the placenta to pass through the inferior vena cava into the right atrium, where it is then directed across the foramen ovale into the left atrium. The oxygenated blood then flows into the left ventricle and directly out to the body of the fetus via the aorta. At birth, the umbilical blood flow is interrupted, stopping the high-volume flow from the placenta. In addition, the alveoli and pulmonary vessels open when the infant takes its first breath, dropping the resistance in the lungs and allowing more flow into the left atrium from the lungs. This reverse in pressure difference between the atria pushes the flexible septum primum against the ridged septum secundum and closes off the foramen ovale and ostium secundum, resulting in the complete septation of the heart chambers [1] (Fig. 3.6). For more information on defects and repairs of the foramen ovale, see Chap. 37.

3.5 Proepicardial Organ and Coronary Artery Development

The last major contributor to vertebrate heart development discussed in this chapter is the PEO [15, 21, 22]. Prior to heart looping, the primary heart tube consists of endocardium, cardiac jelly, and myocardium. It is not until the start of heart looping that epicardial cells from the PEO surround the myocardium, forming the fourth layer of the primary heart tube called the epicardium [15, 53] (Fig. 3.3). This population of cells will eventually give rise to the coronary vasculature. A neural crest origin of the coronary vessels was originally hypothesized, but recent lineage tracing studies have shown that the neural crest gives rise to cells of the tunica media of the aortic and pulmonary trunks but not the coronary arteries [29, 54]. These experiments eventually showed that the coronary vasculature is derived from the PEO, a nest of cells in the dorsal mesocardium of the sinus venosus or septum transversum. These cells, which are derived from an independent population of splanchnopleuric mesoderm cells, migrate onto the primary heart tube (Fig. 3.3) between days 22 and 28 of human development, just as the heart initiates looping [1, 30]. Prior to migration, these cells are collectively called the PEO (or proepicardium). Interestingly, three lineages of the coronary vessel cells (smooth muscle, endothelial, and connective tissue cells) are segregated in the PEO prior to migration into the heart tube [29, 55]. These cells will coalesce to form coronary vessels de novo via the process of vasculogenesis [56]. Recently, it has also been shown that the epicardium provides a factor needed for normal myocardial development and is a source of cells forming the interstitial myocardium and cushion mesenchyme [30, 36]. It is considered that understanding the embryological origin of the vascular system and its molecular regulation may help to explain the varying susceptibility of different components of the vascular system to atherosclerosis [29, 57]. Recently, it has also been suggested that epicardium-derived cells may provide a source of cells for myocardial regeneration after a myocardial infarction [22]. Lastly, among the different stem cell populations identified in the later heart, Isl1-positive cells may be a population of resident cardiovascular stem cells derived from residual SHF cells [12, 58, 59]. Thus, approaches aimed at cardiac repair by manipulation of cardiac progenitor cells will depend on properly understanding how lineage choices are regulated in the SHF and PEO [12, 60].

3.6 Cardiac Maturation

Although the embryonic heart is fully formed and functional by the 11th week of pregnancy, the fetal and neonatal heart continues to grow and mature rapidly, with many clinically relevant changes taking place after birth. During fetal development or from the time after the embryo is completely formed in the first trimester of pregnancy until birth, the heart grows primarily by the process of cell division [61–64]. Within a few weeks after birth, the predominant mechanism of cardiac growth is cell hypertrophy, so that most existing cardiac cells become larger, rather than increasing significantly in number [61–63]. The exact timing of this process and the mechanisms regulating this change are not yet completely elucidated. It has classically been thought that mature cardiac cells lose their ability to divide; however, recent work suggests that limited amounts of cell division do occur in adult hearts that have been damaged by ischemia [65–67].

This finding has led to a renewed interest in understanding the regulation of cell division during cardiac maturation. Additional maturational changes in the fetal and neonatal heart include (1) alterations in the composition of the cardiac myocytes, (2) differences in energy production, and/or (3) maturation of the contractile function. These changes, along with physiologic changes in the transitional circulation, as discussed earlier, significantly affect the treatment of newborns with congenital heart disease, particularly those requiring interventional procedures or cardiac surgery.

The hemodynamic changes associated with birth include significant increases in left ventricular cardiac output to meet the increased metabolic needs of the newborn infant. This improvement in cardiac output occurs despite the fact that the neonatal myocardium has less muscle mass and less cellular organization than the mature myocardium. The newborn myocardium consists of 30 % contractile proteins (mass) and 70 % noncontractile mass (membranes, connective tissues, and organelles). This is in contrast to the adult myocardium which is 60 % contractile mass [63]. The myocardial cells of the fetus are rounded, and both the myocardial cells and myofibrils within them are oriented randomly. As the fetal heart matures, these myofibrils increase in size and number and also orient themselves to the long axis of the rows of cells, which will likely contribute to improved myocardial function [61]. In general, the fetal myocardial cell contains higher amounts of glycogen than the mature myocardium, suggesting an increased dependence on glucose for energy production. In experiments using nonprimate model systems, the fetal myocardium is able to meet metabolic needs with lactate and glucose as the primary fuels [68]. In contrast, the preferred substrate for energy metabolism in the adult heart is long-chain fatty acids, although the adult heart is able to utilize carbohydrates as well [68, 69]. This change is presumably triggered in the first few days or weeks of life by an increase in serum long-chain fatty acids with feeding, yet the timing and clinical impact of this change in ill or nonfeeding neonates with cardiovascular disease remain unknown.

In addition to the changes described above, maturing myocardial cells undergo changes in their expression of many innate contractile proteins, which may be responsible for some of the maturational differences in cardiovascular function. For example, the gradual increase in expression of myosin light chain 2 (MLC 2) in the ventricle from the neonatal period through adolescence is considered to be important in humans. In the fetal ventricle, two myosin light chain forms, MLC 1 and MLC 2, are expressed in equal amounts [63, 70]; MLC 1 is associated with increased contractility and has been documented to increase contractility in isolated muscle from patients with tetralogy of Fallot [71]. After birth, there is a gradual increase in the amount of MLC 2 or the "regulatory" myosin light chain, which has a slower rate of force development, but can be phosphorylated to increase calcium-dependent force development in mature cardiac muscle [63, 72]. There is also variability in actin isoform expression during cardiac development. More specifically, the human fetal heart predominantly expresses cardiac alpha-actin, while the more mature human heart expresses skeletal alpha-actin [61, 73]. Furthermore, actin is responsible for interacting with myosin crossbridges and regulating ATPase activity; work done in the mouse model system suggests that the change to skeletal actin may be one of the mechanisms of enhanced contractility in the mature heart [61, 74, 75]. There are also developmental changes of potential functional significance in the regulatory proteins of the sarcomere. Initially, the fetal heart expresses both alpha- and beta-tropomyosin, a regulatory filament, in nearly equal amounts. After birth, the proportion of beta-tropomyosin decreases and alpha-tropomyosin increases, possibly optimizing diastolic relaxation [61, 76, 77]. In contrast, expression of high levels of beta-tropomyosin in the neonatal heart is associated with early death due to myocardial dysfunction [78]. Lastly, the isoform of the inhibitory troponin, troponin I, changes after birth. The fetal myocardium contains mostly the skeletal isoform of troponin I [61, 79]. After birth, the myocardium begins to express cardiac troponin I, and by approximately 9 months of age, only cardiac troponin I is present [61, 80, 81]. Importantly, cardiac troponin I can be phosphorylated to improve calcium-binding dynamics and contractility, which correlates with improved function in the more mature heart. It is thought that the skeletal form of troponin I may serve to protect the fetal and neonatal myocardium from acidosis [63, 74, 82]. The full impact of these developmental changes in contractile proteins and their effect on cardiac function or perioperative treatment of newborns with heart disease remain unclear at the present time.

Two of the most clinically relevant features of the immature myocardium are its requirement for high levels of extracellular calcium and a decreased sensitivity to beta-adrenergic inotropic agents. The neonatal heart has a decrease in both volume and amount of functionally mature sarcoplasmic reticulum, which is the intercellular storage site for calcium [61]. This paucity of intracellular calcium storage and release via the sarcoplasmic reticulum in the fetal and neonatal myocardium increases the requirement of the fetal myocardium for extracellular calcium, so that exogenous administration of calcium can be used to augment cardiac contractility in the appropriate clinical setting. In addition, neonates and infants are significantly more sensitive to calcium channel-blocking drugs than older children or adults and thus may be at risk for severe depression of myocardial contractility with the administration of these agents [61, 63, 83]. Lastly, although data in humans are limited, there appears to be significantly decreased sensitivity to beta-agonist agents in the immature myocardium and also in older children with con-

genital heart disease [63, 84–86]. This altered sensitivity may be due to: (1) a paucity of receptors, (2) sensitization to endogenous catecholamines at birth or with heart failure, or (3) some combination of these or additional factors. Due to this decreased responsiveness to beta-agonists, there are common requirements for higher doses of beta-agonist inotropic agents in newborns and infants. Note that alternative medications, including phosphodiesterase inhibitors, are often useful adjuncts to improve contractility in newborns with myocardial dysfunction [63].

Although the structure of the heart is complete in the first trimester of pregnancy, cardiac growth and maturation continue to occur in the fetus, newborn, and child. Many of these developmental changes, particularly decreased intracellular calcium stores in the immature sarcoplasmic reticulum and a decreased responsiveness to beta-agonist inotropic agents, significantly impact the care of newborns, infants, and children with congenital heart disease, particularly those requiring surgical intervention early in life.

3.7 Summary of Embryonic Contribution to Heart Development

The contribution of the four major embryonic regions to heart development—FHF, SHF, CNC, and PEO—illustrates the complexity of human heart development. Each of these regions has a unique contribution to the heart, but they ultimately depend on each other for the creation of a fully functional organ. Furthermore, a better understanding of the mechanisms of human heart development will provide clues to the etiology of congenital heart disease. The genetic regulatory mechanisms of these developmental processes are just beginning to be characterized. A molecular review of heart development is outside the scope of this chapter, but several informative molecular heart reviews have been recently published [6, 16, 30, 87, 88]. A better understanding of the embryological origins of the heart, combined with the characterization of the genes that control heart development, will likely lead to many new clinical applications to treat congenital and/or adult heart disease.

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