In vivo and *In vitro* Development of the Chicken Heart

Jacqueline S. McLaughlin

Department of Biology
Pennsylvania State University at Lehigh Valley
8380 Mohr Lane, Fogelsville, PA 18051 USA
(619) 285-5109, jshea@psu.edu

and

Elizabeth R. McCain

Biology Department
Muhlenberg College
2400 Chew Street, Allentown, PA 18104 USA
(610) 821-3255, mccain@muhlenberg.edu

This laboratory experiment is designed to introduce students to the developmental process of the vertebrate heart using the chicken embryo as a model system. Simultaneously, the students are also exposed to the physiology of embryonic blood flow and the electrical circuitry of the developing heart. Described herein is a review of the chicken heart development and a summary of our basic protocol.

Introduction to Heart Development: The development of the heart involves a series of cellular migrations, fusions, and tissue differentiation; i.e., a multitude of morphogenetic mechanisms. The heart of the chicken embryo develops from the fusion of paired precardiac mesodermal tubes located on either side of the developing foregut. Between 25 and 30 hours of incubation, the paired heart vesicles begin to fuse at the anterior (head) end and continue to fuse posteriorly to form one continuous tube. After fusion is complete, the heart tube is ventral to the foregut and has four distinct regions that can be identified from anterior to posterior: **bulbus cordis, ventricle, atrium, and sinus venosus.** Blood flows anteriorly, from the sinus venosus to the bulbus cordis. At approximately 33 hours, the heart tube bends to form an "S" shape, with the prominent ventricle bulging to the right. By 48 hours, the heart has folded upon itself, forming a single loop. This moves the sinus venosus and atrium to a position anterior and dorsal to the ventricle and the bulbus cordis. The ventricle is U-shaped and in the medial ventral position. Now, the blood flows posteriorly and then makes a sharp turn to flow anteriorly. In the 72-hour embryo, the atrium has begun to expand to the left in preparation of the division into the right and left atria.

Although the heart still has two chambers at this time, communication between the sinus venosus and the atrium is via the right side of the atrium. This is the first step towards the sinus venosus becoming part of the future right atrium.

The heart begins to beat just after the paired heart rudiments begin to fuse, immediately before the bulbus cordis forms. Once the heart tubes have completely fused, the sinus venosus becomes the embryonic pacemaker. Eventually, when the atrium and ventricle each divide into a pair of chambers

and a typical four-chambered heart is present, the sinus venosus is incorporated into the right atrium where it gives rise to the sinoatrial node, a pacemaker.

Laboratory Procedures: Students begin this exercise by identifying the anatomy of 48-hour or 60-hour and 72-hour chicken embryos. This *in vivo* technique requires windowing the eggs using a modified procedure from Cruz (1993). The students also determine the *in vivo* heart rate and direction of blood flow through the heart. Subsequently, the embryos are explanted (Cruz, 1993) to observe more closely the *in vitro* heart contractility and blood flow. In order to study the intrinsic ability of heart contraction, the hearts are then surgically removed using iris microdissecting scissors or handmade microknives (Tyler, 1994). The explanted hearts are further dissected into the four chambers (**sinus venosus, atrium, ventricle, bulbus cordis**) to allow identification of the embryonic pacemaker. Once the students are efficient with the above procedures, they are required to design an experiment evaluating the effects of caffeine or gin on the *in vivo* or *in vitro* heart rate. A written protocol, with a clearly defined hypothesis, is required of the students before experimentation. Following data analysis, a full length lab report is submitted.

Chick Usage: Chick embryos are picked up from a local chicken farm the day they will be used. Eggs are kept at 37^{O}C in a styrofoam incubator (cost, $50 US) from Carolina Biological.

The students appreciate studying a more advanced vertebrate organ system, carrying out dissections, using fine tools, and designing their own experiments using common teratogens. Future work includes having students design experiments which investigate the long-term effects of various teratogens on chicken heart development.

Literature Cited

Tyler, M.S. 1994. Developmental Biology: A Guide for Experimental Study. Sinauer Associates, Inc., Massachusetts, 172 pages.

Cruz, Y.P. 1993. Laboratory Exercises in Developmental Biology. Academic Press, California, 241 pages.

Reprinted From: McLaughlin, S. J. and E. R. McCain. 1998. *In vivo* and *in vitro* development of the chicken heart. Pages 331-332, *in* Tested studies for laboratory teaching, Volume 19 (S. J. Karcher, Editor). Proceedings of the 19th Workshop/Conference of the Association for Biology Laboratory Education (ABLE), 365 pages.