DEVELOPING A MODEL THREE-DIMENSIONAL ANIMATION OF

EMBRYONIC HEART DEVELOPMENT

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DEDICATION

I would like to thank the members of my Graduate Committee, especially Dr. Rhonda

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also like to thank Dr. Jens Fielitz, Dr. James Richardson, and all of the members of the

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DEVELOPING A MODEL THREE-DIMENSIONAL ANIMATION OF

EMBRYONIC HEART DEVELOPMENT

by

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THESIS

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DEVELOPING A MODEL THREE-DIMENSIONAL ANIMATION OF

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Three-dimensional animations are often effective in explaining complex phenomena, but altering the finished productions can be cumbersome and costly. The goal of this thesis was to develop a technique for easily altering 3D animations using 2D methods. In conjunction with the Olson Laboratory at the University of Texas Southwestern Medical Center, I created two animations dealing with heart development, based on one 3D animation model. The animations focused on cardiac morphogenesis and transcription factor expression. The purpose of this project was to not only visually communicate cardiac morphogenesis and the expression patterns of transcription factors, but to create a 3D model that can be used as a template for any further visualization of heart development research.

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Congenital Heart Disease – Heart disease that presents itself at birth.

Embryogenesis – The growth and development of an embryo.

Morphogenesis – Various processes occurring during development by which the form of the body and its organs is established.

Transcription – In protein synthesis, the process of copying the genetic information from the DNA in the chromosomes to the ribosomes, cell organelles in the cytoplasm that are the site of protein synthesis.

Transcription factor – A contributing cause (a gene) for the process of transcription.

CHAPTER ONE
Introduction

<u>Goal</u>

Three-dimensional animations are often effective in explaining complex phenomena, but altering the finished productions can be cumbersome and costly. The goal of this thesis was to develop a technique for easily altering 3D animations using 2D methods. To reflect current research, two-dimensional animation methods can be employed to quickly build or update the 3D animation. This technique allows researchers to use the same model to visualize multiple topics, such as cardiac morphogenesis or the expression patterns of various transcription factors. Visualizing the developing heart is nearly impossible with two-dimensional illustrations, so the ability to create quick three-dimensional animations, using 2D methods, is vital to researchers.

In order to accomplish this goal, I set three main objectives. The first was to research possible subject matters with the Olson Laboratory at the University of Texas Southwestern Medical Center at Dallas to determine what two topics would be the basis of the animations. The second was to create illustrations and three-dimensional models that accurately represent the anatomical stages of heart development. Last, I proposed to create two animations that met the needs for a specific audience using two-dimensional methods.

Two animations were chosen to demonstrate how one "model" animation is used to quickly create a new animation. The first depicts cardiac development with color coding of morphologically related regions. Using the same basic model, the second

animation maps the expression patterns of eight transcription factors to their locations within the developing heart.

This project was created in conjunction with Eric Olson, Ph.D., Rhonda Bassel-Duby, Ph.D., and the Olson Laboratory at the University of Texas Southwestern Medical Center at Dallas. The project targets scientists involved in heart research, including molecular biologists, cardiologists, graduate students, and post-doctoral fellows. This unique visualization enhances understanding of the process of heart development and the mechanisms involved, and will hopefully stimulate further avenues of investigation.

Background Information

Cardiovascular disease is the leading cause of death in the United States, accounting for over 30% of adult deaths (McConnell, 94, 199). Similarly, the primary cause of non-infectious death in children is attributed to congenital heart disease (CHD) (McConnell, 215 and Srivastava & Olson, 221). By understanding the mechanisms that control the formation of the heart during embryogenesis, researchers hope to reduce the risk of cardiovascular disease and CHD. Cardiac muscle cells express distinct sets of genes controlled by different combinations of transcription factors and extracellular signals. These transcription factors and extracellular signals affect genes, and are responsible for controlling programs of cell differentiation and morphogenesis during development.

Morphology of Cardiac Development

Human cardiac development occurs during weeks 4-8[1], post conception. However, a primitive placental circulation is already established by week 2. These heart precursors form a crescent, or cardiac primordium, that is specified to form particular segments of the developing heart (Srivastava & Olson, 223). Conotruncal, ventricular, and atrial regions are apparent at this stage (Figure 1).
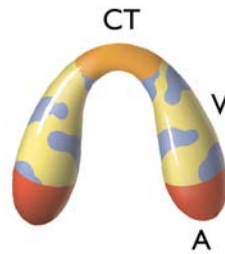

Figure 1 – The Cardiac Crescent. Conotruncal (CT), ventricular (V), and atrial (A) regions are apparent at this stage.

By week 3 of development, the cardiac crescent forms a linear heart tube. The linear heart tube is specified into aortic sac, conotruncal, right and left ventricular, and right and left atrial segments (Srivastava & Olson, 223) (Figure 2).

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[1] Human developmental age is indicated here, however, research is based on days of mouse heart development.
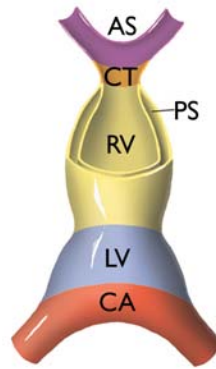
Figure 2 – The Linear Heart Tube. The aortic sac (AS), conotruncus (CT), primordial sac (PS), right (RV) and left (LV) ventricles and common atrium (CA) are apparent at this stage.

By week 4, the chambers of the heart balloon outward and the linear heart tube begins to loop. The ventriclular regions descend and loop rightward as the atria loop posteriorly, moving behind the ventricles. The common atrium differentiates into the right and left atria at this stage (Srivastava & Olson, 223) (Figure 3).
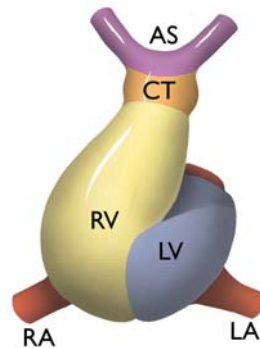

Figure 3 – The Looping Heart. The aortic sac (AS), conotruncus (CT), right ventricle (RV), left ventricle (LV), right atrium (RA), and left atrium (LA) are apparent at this stage.

After the looping of the heart, septation occurs and distinct chambers are formed. The atria move superiorly and assume their position behind the conotruncus. Endocardial

cushions, which will later differentiate into heart valves, are evident. Trebeculation

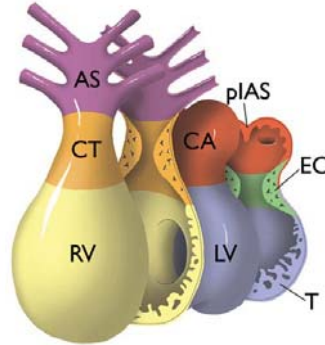begins to form in the right and left ventricles (Figure 4).

Figure 4 – Chamber Formation and Septation. The aortic sac (AS), conotruncus (CT), right ventricle
(RV), left ventricle (LV), right atrium (RA), left atrium (LA), primary atrial septation (pIAS), endocardial
cushion (EC), and trebeculation (T) are apparent at this stage.

At week 8, the embryonic heart is fully formed and functioning. The aortic sac

forms the aorta and pulmonary artery. The aortic and pulmonary trunks join with arteries

from branchial arches III, IV, and VI to form the subclavian arteries, common carotid

arteries, and ductus arteriosus. The atria move anteriorly, arriving in their final position

superior to the ventricles. The ventricular and atrial chambers are fully septated and the

valves have developed from the endocardial cushion (Wilson, E8,4 and Srivastava &
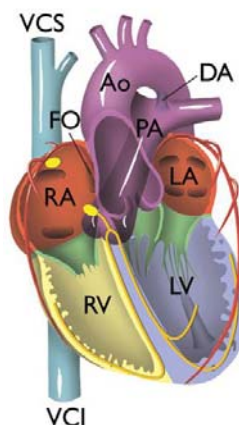
Olson, 223) (Figure 5).

Figure 5 – The Mature Heart. The ventricles (LV, RV), atria (LA, RA), aorta (Ao), pulmonary artery (PA), ductus arteriosus (DA), vena cava (VCS, VCI), and foramen ovale (FO) are shown.

Expression Patterns of Transcription Factors

The Olson Laboratory's primary focus is on muscle cells and how they adopt certain fates. In particular, the lab researches how cardiac muscle cells express genes that are controlled by certain combinations of transcription factors and extracellular signals. By isolating the transcription factors that control cardiac development, researchers hope to better understand how the heart develops and what mechanisms are involved. Studying mutations of these transcription factors, and the resulting heart defects, will hopefully lead to a better understanding of the complex process of embryogenesis, as well as a possible cure for cardiovascular disease and CHD.

Among the transcription factors studied by the Olson Laboratory are HAND1 (Figure 6), HAND2, Myocardin, Nkx2.5, Tbx5, GATA4, HRT1, and HRT2. The expression patterns of these factors are the focus of the second animation. Dr. Olson's lab works most extensively with four of the above transcription factors, HAND1, HAND2, Nkx2.5, and Myocardin, which are crucial to proper cardiac cell differentiation

and morphogenesis. HAND1 is expressed predominantly in the primitive left

ventricular segment of the linear heart tube. HAND2 is expressed predominantly in the

primitive right ventricular segment of the heart tube. Nkx2.5 is thought to act upstream

of HAND1, and to be expressed in the left ventricular portion of the cardiac crescent.

Myocardin, a novel gene recently discovered by Dr. Olson, has a similar expression

pattern to Nkx2.5. Its expression is initiated in the cardiac crescent at the time of

cardiogenic specification, and continued to be found in the atrial and ventricular

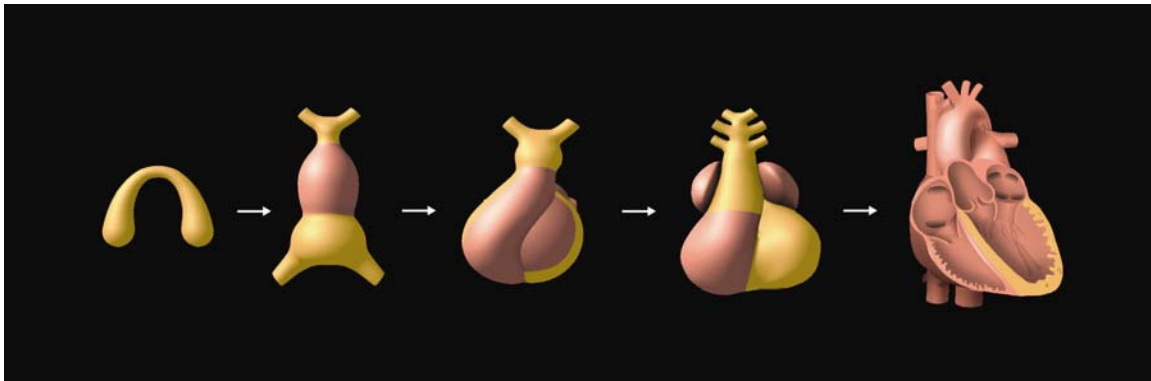chambers of the heart during later development (Srivastava & Olson, 223-225).

Figure 6 – HAND1 expression.

Significance of Project

The purpose of this project was to not only visually communicate cardiac

morphogenesis and the expression patterns of transcription factors, but to create a 3D

model that could be used as a template for any further visualization of heart development

research. The project aimed to construct an animation that is easily updated or changed

to reflect the rapid advancements of scientific discovery. The production process itself was vital to developing an animation that meets these requirements.

Limitations

Originally, I planned to create nine independent videos from one 3D animation model, including an individual video for each of the eight transcription factors discussed. Eventually, the number of videos was limited to two; one video depicting cardiac morphogenesis, and one video that included the expression patterns of all eight transcription factors. The 3D aspects of the video were limited to very basic modeling and animation, with the majority of detailed animation effects reserved for the 2D editing software.

Production Methods

A storyboard of the animations was created, along with detailed sketches of the developing embryo. Sketches included the locations of cardiac precursors, tissue migration, and expression of transcription factors. Six 3D models were made and animated in 3ds max™ 6. Two-dimensional stills were taken from the 3D animation and morphed using WinMorph™ 3.01. Two-dimensional animation effects were created in Adobe After Effects® 6.5. The final animations were edited in Adobe Premiere® Pro 1.5 and recorded onto CD-ROM.

<u>Evaluation</u>

To evaluate the project, an informal survey was taken by Post-Doctoral Fellows and Project Investigators in the Olson Laboratory and the Department of Cardiology. The audience viewed the two final animations and answered a short questionnaire. The questionnaire was designed to test the animations' effectiveness on two target audiences.

CHAPTER TWO
Review of the Literature

SCIENTIFIC OVERVIEW

Introduction

      Recent studies show that many heart abnormalities have direct links to mutations in certain developmental transcription factors. The sequence of cardiac morphogenesis, from the cardiac crescent to the mature heart, has been established for years. However, researchers have only just uncovered the mechanisms that control this developmental process, and they are identifying the control genes involved.

The Role of Genetics in Congenital Heart Disease

      The origins of congenital heart disease (CHD) are still not completely understood. Researchers believe that a pattern of variable penetrance and variable phenotype is involved in CHD, although the genetic blueprint is extremely complex (Srivastava & Olson, 221). Eight main transcription factors have been identified in this process, and are the focus of this project. The expression patterns of each depict a clear, if not all encompassing, picture of the genes' involvement in cardiac morphogenesis.

HAND1

      HAND1 (*h*eart, *a*utonomic nervous system, *n*eural crest-*d*erived cell type) (Harvey & Rosenthal, 146) is expressed predominantly in the primitive left ventricular segment of the linear heart tube (Figure 7). The factor is thought to have a role in

development of the left ventricle, along with Nkx2.5, mentioned below (Srivastava &
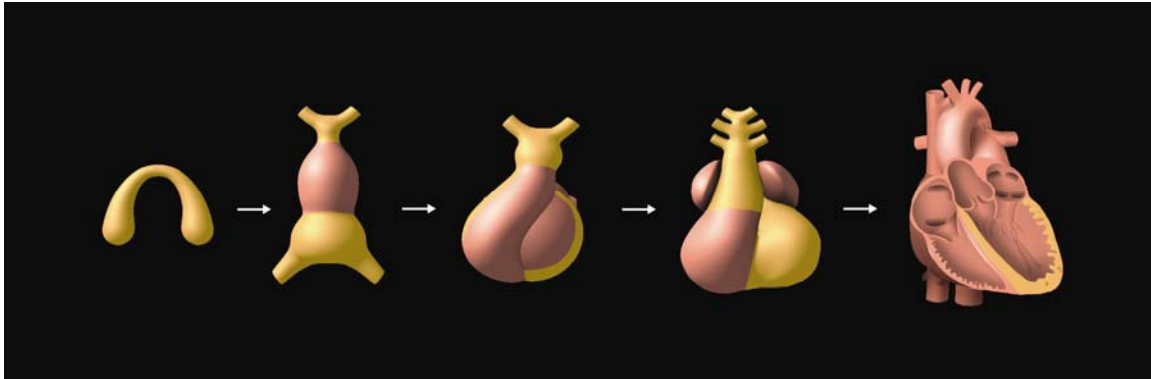
Olson, 148-153).

Figure 7 – HAND1 expression.

HAND2

　　While HAND1 is primarily involved in development of the left ventricle, HAND2

is similarly involved in the right ventricle (Figure 8). The transcription factor is

expressed primarily in the right ventricle of the developing heart, and mutations of

HAND2 result in hypoplasia of the right ventricle (Srivastava & Olson, 223). Both

HAND1 and HAND2 have also been implicated in development of the aortic sac, aortic

arch arteries, and other neural crest-derived structures (Harvey & Rosenthal, 148-153).
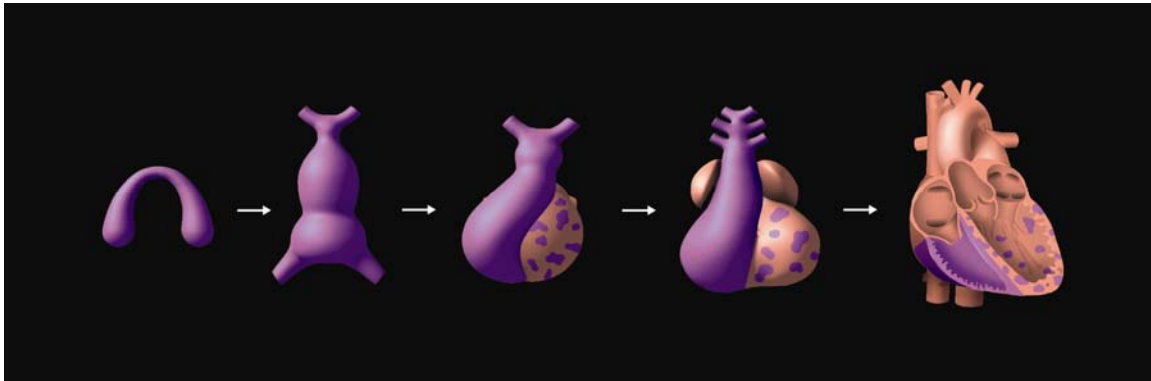
Figure 8 – HAND2 expression.

Myocardin

Myocardin is a novel gene recently discovered by Dr. Olson, and little has been published on its involvement in heart development. Its expression is initiated in the cardiac crescent at the time of cardiogenic specification, and continued to be found in the atrial and ventricular chambers of the heart during later development (Figure 9).
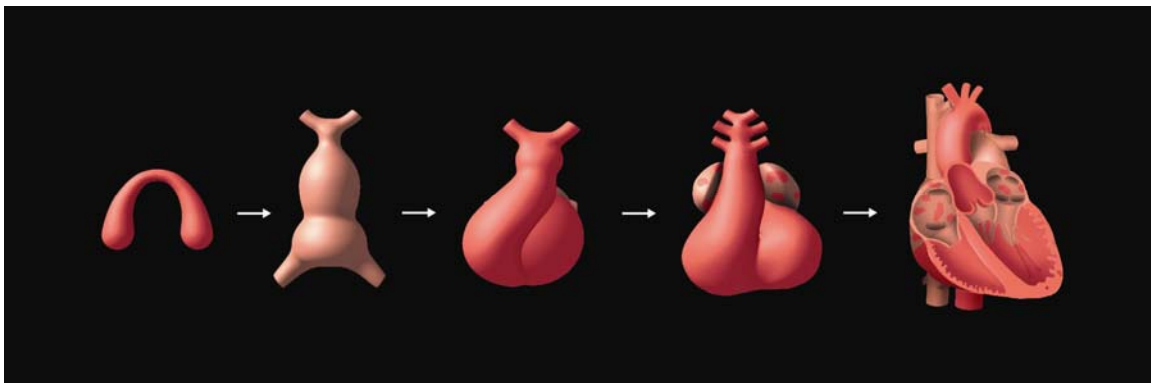

Figure 9 – Myocardin expression.

Nkx2.5

Nkx2.5 has a similar expression pattern to myocardin. The transcription factor is thought to act upstream of HAND1, and to be expressed in the left ventricular portion of

the cardiac crescent (Figure 10). Nkx2.5 mutations are implicated in atrial septal

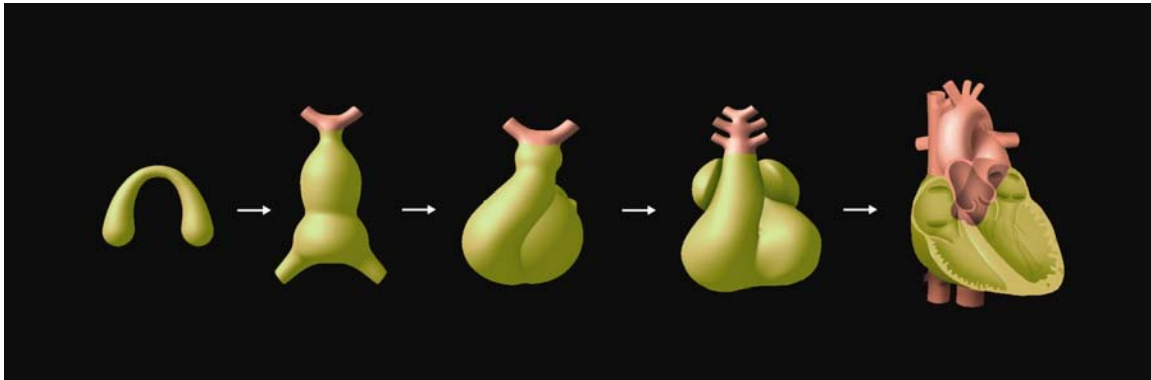defects, cardiac conduction abnormalities, and the tetralogy of Fallot (Srivastava &

Olson, 225).

Figure 10 – Nkx2.5 expression.

Tbx5

Tbx5 mutations lead to ventricular and atrial septal defects (Srivastava & Olson,

225). Tbx5 is expressed in the left ventricular region of the heart, as well as both the
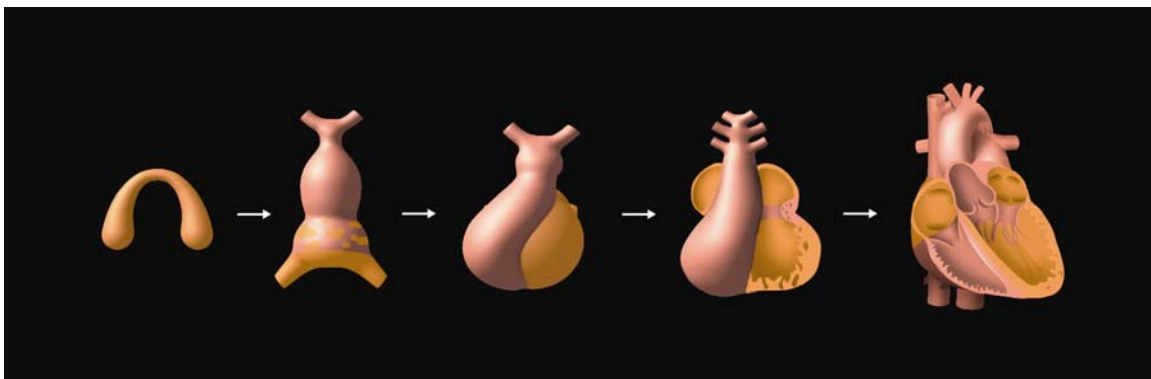
right and left atrial regions (Figure 11).

Figure 11 – Tbx5 expression.

GATA4

GATA4 (Figure 12) expression first appears in the ventricular and atrial segments of the linear heart tube, and is essential for ventral mophogenesis and formation of the heart tube (Harvey & Rosenthal, 303). Mutations of GATA4 lead to cardiac bifida (Srivastava & Olson, 225).
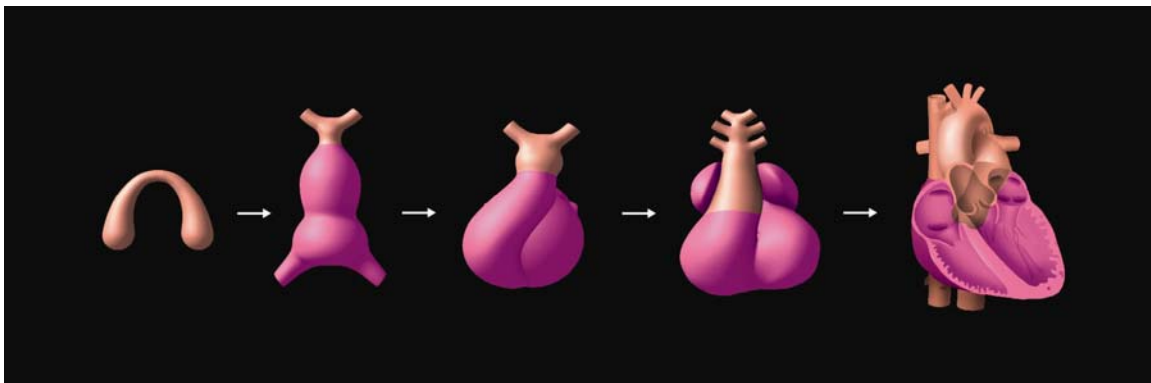

Figure 12 – GATA4 expression.

HRT1

HRT1 is expressed in the atria, as well as the endocardial cushion of the conotruncus (Figure 13). The Department of Molecular Biology at UT Southwestern has located the expression pattern of this transcription factor, however, very little has been published regarding the direct role HRT1 plays in heart formation.
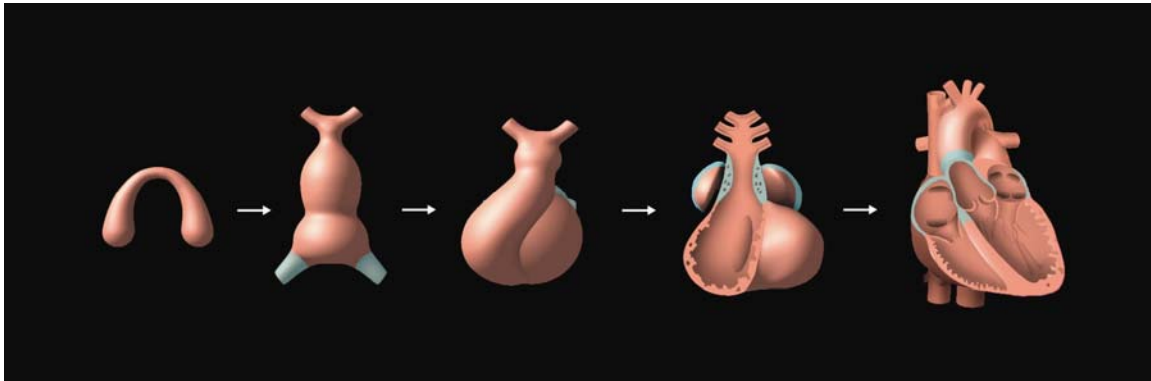
Figure 13 – HRT1 expression.

HRT2

Expression of HRT2 is observed in the ventricles and endocardial cushions. Like HRT1, there is little published on the role of HRT2 in heart development (Figure 14).
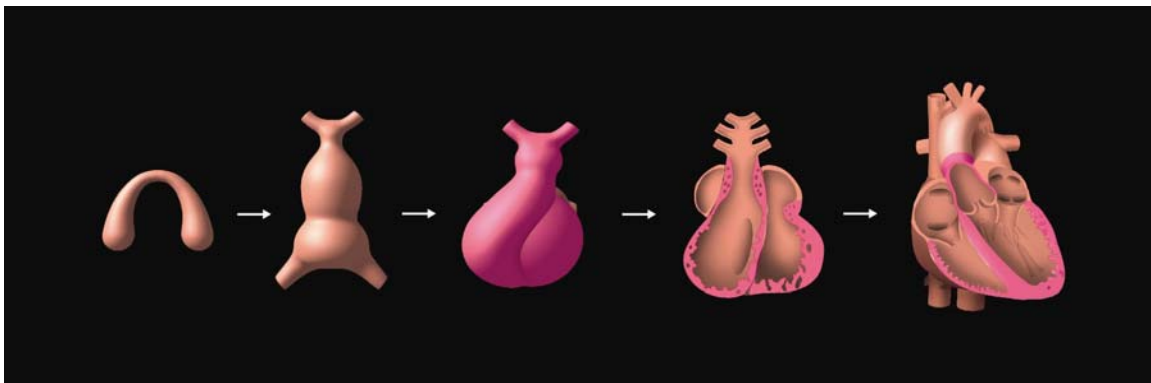

Figure 14 – HRT2 expression.

EXISTING LITERATURE

The majority of visuals on the subject of heart development are two-dimensional, schematic illustrations. Most of the illustrations focus on three or four specific developmental stages, and leave the intermediate stages to the audience's imagination. Though illustrations of cardiac morphology are numerous, few of them focus on the relationship between transcription factor expression and cardiac development. Similar to the basic morphology illustrations, these were also basic two-dimensional drawings (Figure 15).
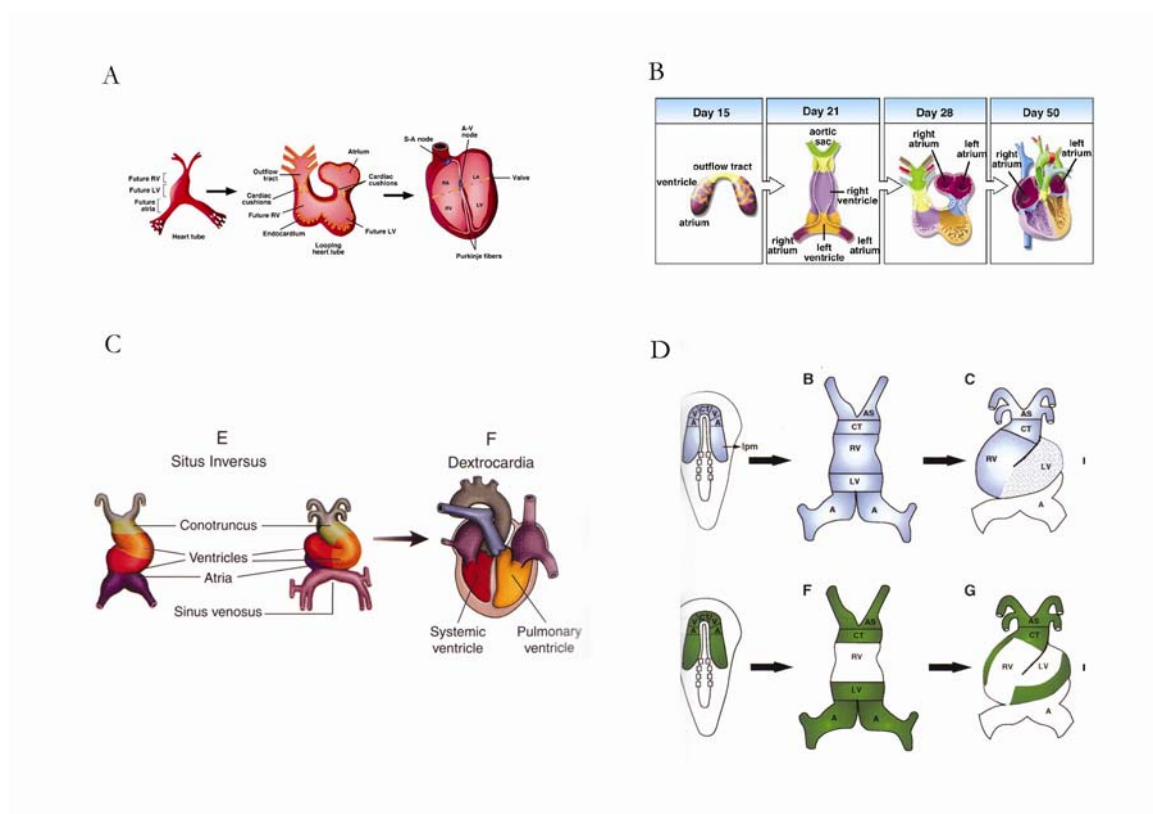

Figure 15 – Examples of existing heart development illustrations. A (Nat Med © 2004), B (Nature © 2000), C and D (Harvey & Rosenthal © 1999)

Histological sections and scanning electron micrographs are the most accurate representations of cardiac development, however, they can be confusing. Likewise, they also tend to depict certain stages from only one angle. The difficulty in conceptualizing these images can overshadow their accuracy (Figure 16).
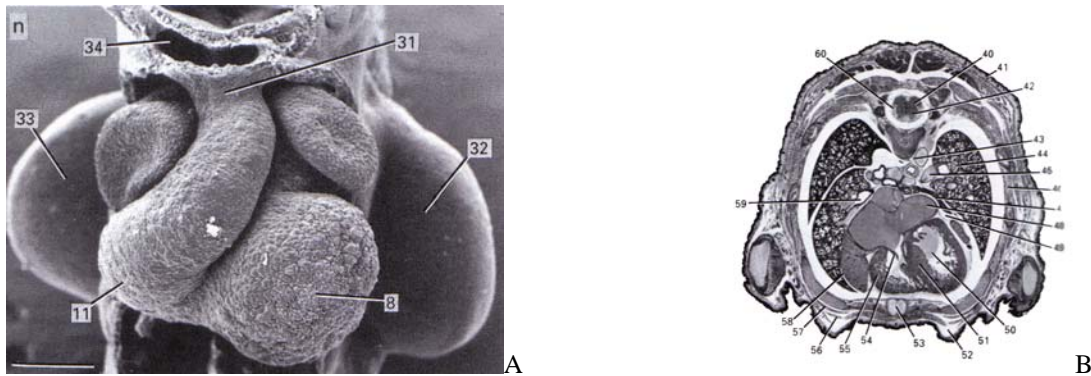

Figure 16 – Electron micrograph (A) and histological section (B) views of heart development. (Kaufman)

Three-dimensional animation is the best way to fully visualize the developing heart. Numerous heart animations are easily accessed over the internet, however, the majority of these were simulations of beating, mature hearts. A few animations depicted heart development, however the majority of these were Flash animations of two-dimensional drawings. Some animations were complex three-dimensional models, but even these portrayed the heart from only one angle, or only showed partial development. None of the animations displayed a 3D model morphing fluidly from cardiac crescent to mature heart. Nor did they show the side and rear angles of the developing heart.

Conclusion

       After reviewing the literature, I realized how few resources there are pertaining to genetic involvement in cardiac morphogenesis. I wanted to create a 3D animation that not only depicted cardiac morphogenesis in a way never shown before, but that also could be easily updated as new discoveries are made. The prevalence of cardiovascular disease and CHD in our country stress the importance of visualizing possible mechanisms of heart development.

CHAPTER THREE
Methodology

Introduction

Preparation and development were essential to successful completion of this project. In order to create a basic model of heart development that could be easily altered or reused, it was crucial to plan carefully. The methods chosen were carefully thought out, and selected for their value to the final product.

Concept and Planning

From the beginning, the concept was to create a 3D animation of embryonic heart development, however the particulars were unknown. Members of the Olson Laboratory decided what elements of heart embryogenesis were most critical to the lab. Eventually, two main themes were chosen; the first being basic heart morphology and the second being transcription factor involvement in cardiac development.

Certain decisions were made in order to meet the needs of the target audience, namely project investigators and graduate students in the Molecular Biology and Cardiology labs. The animations had to be accurate and technical enough for an advanced audience yet had to indicate enough basic anatomy to serve as a teaching mechanism for students. The first of the two animations appeals to a less knowledgeable audience, while the second animation deals with the advanced concept of transcription factor expression.

The morphology of the first animation was relatively easy to demonstrate, but deciding how to represent the transcription factors was more difficult. Eight transcription

factors, which are researched in the Olson Laboratory, were included in the animation. A discussion arose about how best to show the expression of these factors. Originally, we planned to create eight small animations, each depicting the expression of one factor. Finally, it was decided to show all eight factors in one animation in order to emphasize their interactions.

Planning the animation involved devising a method to make it easy to update. It was decided early on to create as much of the animation as possible with two-dimensional techniques. A limited number of three-dimensional models were made, and any rendering from 3ds max™ was kept to a minimum. Two-dimensional stills replace three-dimensional models in much of the video. Likewise, the colorization was achieved in Adobe Photoshop® and Adobe After Effects®, versus creating texture maps in 3ds max™. Every effort was made to avoid the necessity of returning to 3ds max™ in order to make changes.

Hardware

The animation was created on a Dell™ Intel Pentium processor with 930 MHz and 512 MB of Ram. The operating system used was Microsoft® Windows® XP Professional, version 2002.

Software

3ds max™ 6 was used to create the 3D models and to render the animation sequences. WinMorph™ 3.01 was used for two-dimensional morphing. Colorization of

the models was achieved in Adobe Photoshop® 7.0 and animated two-dimensionally in Adobe After Effects® 6.5. Adobe Premiere® Pro 1.5 was used for editing and labeling the animation.

Storyboards and Sketches

Two storyboards were created to detail the main actions and events of the two animations (Appendix A). They included notes on how the animation would progress, and what labels and information would be included. Rough sketches of five main developmental stages were shown to several content experts for their opinions and feedback (Appendix B). Numerous revisions of the illustrations were completed before the sketches were finally approved.

The fourth heart stage, which illustrates the chamber formation and septation after the heart loops, proved to be the most difficult stage to perfect. Many revisions were made to accurately depict what occurs at this time. Electron micrographs from Kaufman's *The Atlas of Mouse Development* helped to improve the sketches.

3D Models

Because of the lack of complete information on how tissue migration occurs during morphogenesis, and the limitations of 3ds max™, it was impossible to make only one 3D model and morph it from beginning to end of morphogenesis. Several models were needed to give the illusion of "morphing".

The original plan was to create four 3D models in 3ds max™. Because of

morphing difficulties, six were eventually made. Models were built using the subdivision

surface technique of modeling. Basic shapes were converted to editable poly mode and

altered in order to create the six heart stages (Figure 17). The 3D models were approved
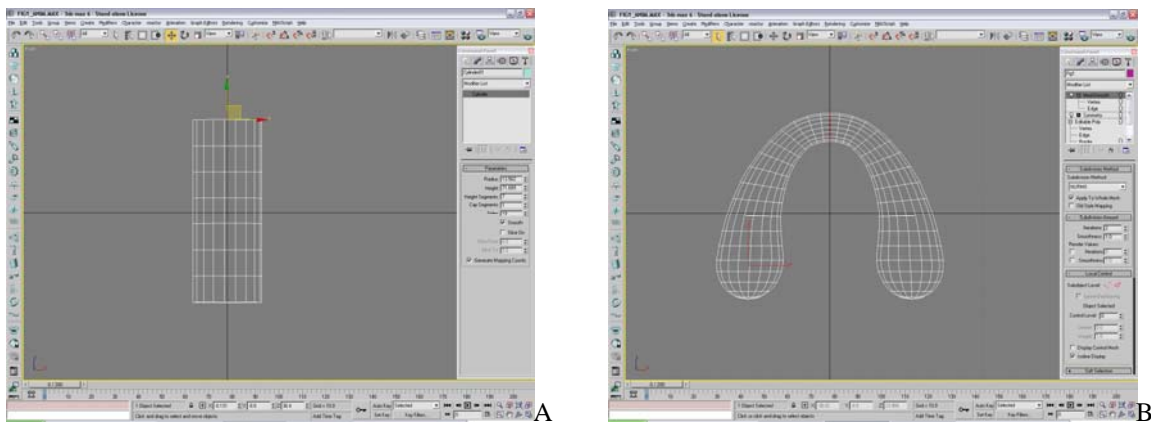
by my thesis committee before proceeding.

Figure 17 – Subdivision Surface Modeling. Shape A was bent and extruded to create shape B.

A smooth surface was chosen for the models by applying a Mesh Smooth

modifier with 2 iterations. A neutral ambient and diffuse color was chosen, and set to a

specular level of 56, with a glossiness level of 35. No bump map, or other surface map,

was added.

Lights and cameras were added to the scenes. The camera was animated around a

circle spline in order to make the models appear to rotate. Two-hundred frames were

used to show each model rotate.

<u>Rendering</u>

Each animation was rendered at 720 x 480, with an Image Aspect of 1.333 and a

Pixel Aspect of 0.9. Animations were rendered Super Black in order to make later

adjustments during the editing phase. The animations were rendered out as AVI files and

brought into Adobe Premiere® for editing. The Super Black setting was especially

useful in matching up the AVI files from 3ds max™, WinMorph™, and Adobe After

Effects® so that they ran seamlessly.

<u>Morphing</u>

Morphing the heart from its earliest stage, the cardiac crescent, to the mature heart

proved to be the most difficult element of the project. The first attempts to morph the

heart using 3ds max™ failed. I tried to use the Morpher Modifier in 3ds max™, but after

many failed attempts, realized that the models were too complicated for this affect. I

abandoned the notion of morphing in a 3D program, and moved on to 2D morphing

programs.

WinMorph™ 3.01 was eventually used to morph the heart two-dimensionally.

Still frames of the six heart stages were taken, and brought into WinMorph™ as JPEGs.

Splines were used to morph one image into the next, and the appearance of three-

dimensional morphing was achieved (Figure 18). The morphs were rendered out as AVI

files, at 24.0 frames per second, with accurate warp precision. Smart Overlay was used,

and the animations were compressed using Cinepak Codec by Radius at 100% quality.
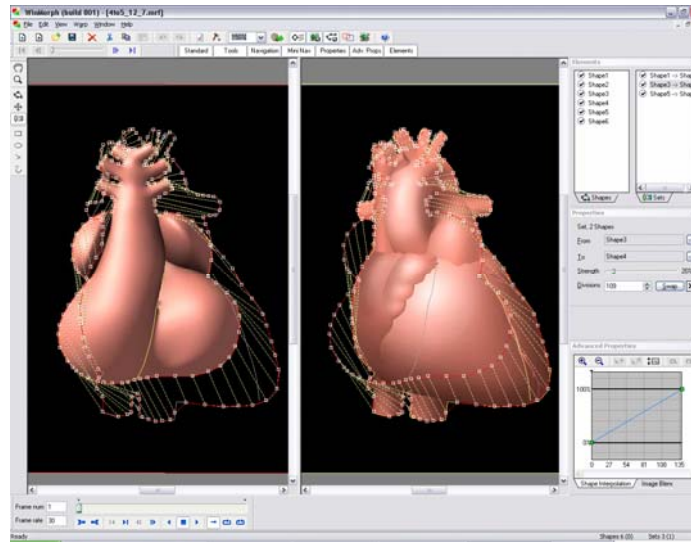
Figure 18 – Morphing in WinMorph™.

Six morphing animations were made, and edited together in Adobe Premiere®, to give the illusion of the heart morphing from start to finish. The WinMorph™ software was not as accurate as it could have been, leaving unclean edges during the morphing process. Because of this, some elements of the morphing sequence were cleaned up using masks in Adobe After Effects® (Figure 19).
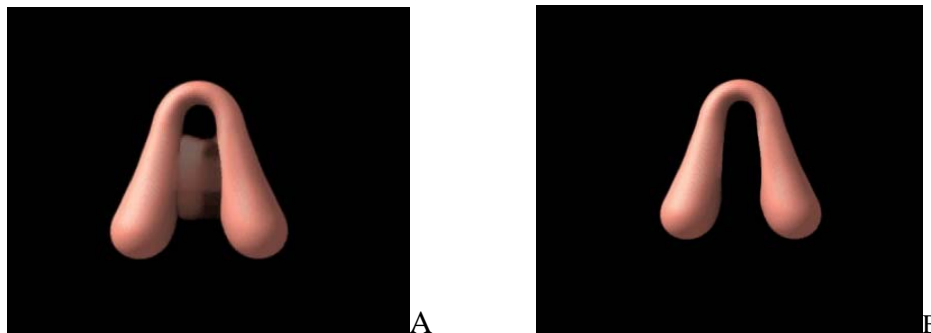

A   B

Figure 19 – WinMorph™ left uneven edges and shadowed images (A) that were later cleaned up in After Effects®.

2D Animation Effects

   Adobe After Effects® was used, not only to perfect the morphing sequence, but also to colorize and animate certain parts of the project. JPEGs of the models were brought into Adobe Photoshop® and colorized to show highlighted areas for labeling (Figure 20). The Photoshop documents were then brought directly into After Effects® as compositions, and the layers were maintained. By adjusting the opacity of colored layers at certain key points, the models were animated and ready for labeling.

   AVI files were rendered from After Effects®, using Video for Windows® at 15 frames per second, and Microsoft® DV compression. The composition settings were set at 720 x 480. The AVI files were then brought into Adobe Premiere®.
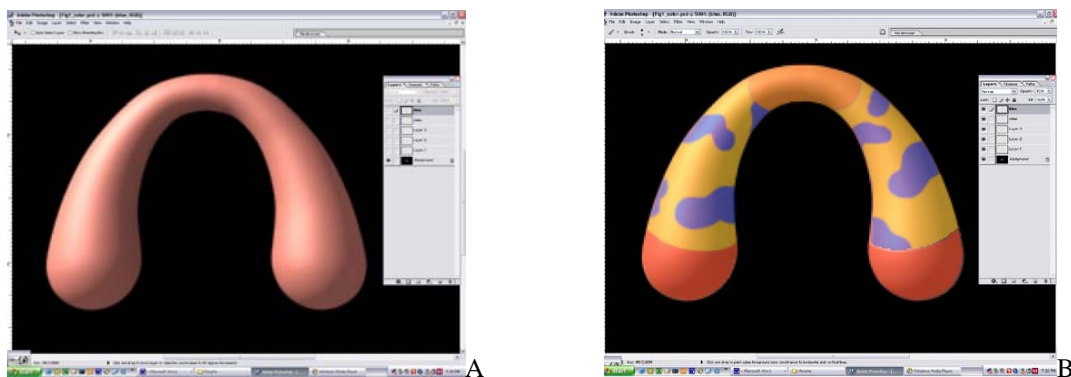
Figure 20 – Still shots from 3ds max™ (A) were taken into Photoshop® and colorized. The Photoshop® layers (B) were then animated two-dimensionally in After Effects®.

Editing

   Adobe Premiere® was used for editing and labeling the animation. Labels were inserted to correspond to the animated highlights added in After Effects®. AVI files from 3ds max™, WinMorph™, and After Effects® were brought together, along with

some JPEG stills. The components were combined in such a way as to present a seamless animation. Fades were added, along with a title page and copyright information.

Although AVI files were chosen for rendering because of quality, Quicktime files were chosen for the final product due to file size considerations. The Quicktime animations were rendered out of Premiere® with Animation compression at 50% quality. The animations were rendered at 720 x 480 and 0.9 D1/DV NTSC. Recompression was chosen.

CD-ROM Creation

The Quicktime files were burned onto CD using Roxio™. Three separate CDs were made, one with the Heart Development Morpholgy animation, one with the Transcription Factor Expression animation, and one with the morphing sequence only.

Updating and Reusing the Videos

The 3D model animation can be easily updated by anyone knowledgeable about animation techniques. The first step is to bring the 3D model AVI files into After Effects®. Next, one would create new Photoshop® documents relating to the updates or new subject matters, and bring those files into After Effects® as well. The Photoshop® files are then manipulated and animated two-dimensionally on top of the 3D animation. Finally, the new animation is rendered out of After Effects®. There is no need to go back

into 3ds max™, which sharply cuts the time and money involved in animation production.

Conclusion

During the production process, every attempt was made to keep the 3D model animation generic and simple. Whenever possible, attempts were made to use two-dimensional methods, and to reduce the necessity of returning to the 3D program used for the model. By keeping the model animation simple, multiple animations could be created from it.

CHAPTER FOUR
Results

Eleven researchers participated in an informal survey of the two heart animations (Appendix C). Six members of the Department of Molecular Biology and five members of the Department of Cardiology viewed the two animations, and answered a ten-question survey. The questionnaire assessed the effectiveness of the animations, and the overall usefulness of three-dimensional animations versus two-dimensional illustrations.

On a scale of one to five, five being the most effective, question one asked, "*How effective do you believe the first video (cardiac morphogenesis) is in depicting how the heart develops?*" Out of the eleven people, three gave a score of 5, six gave a score of 4, and two gave a score of 3 (Table 1).
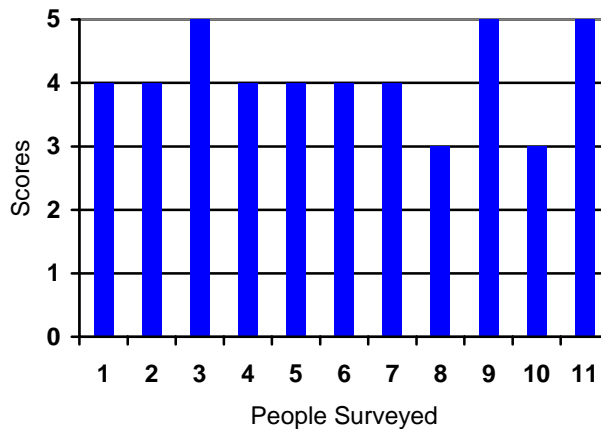
Table 1 - Question one - "How effective do you believe the first video (cardiac morphogenesis) is in depicting how the heart develops?"

The second question asked, "*How effective do you believe the second video (transcription factors) is in depicting the expression of transcription factors in the*

28

developing heart?" Of the eleven people surveyed, two gave a score of 5, six gave a

score of 4, and three gave a score of 3 (Table 2).
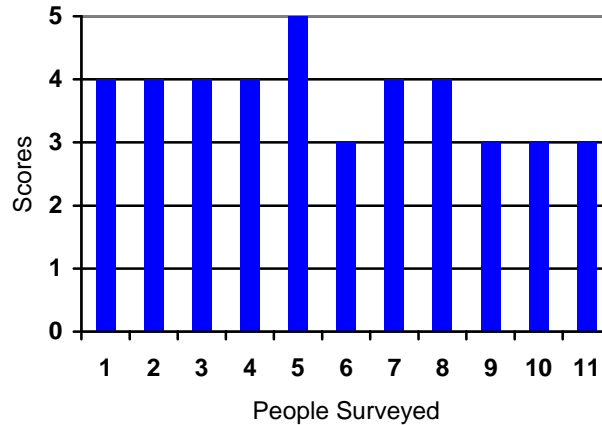
Table 2 – Question Two - "How effective do you believe the second video (transcription factors) is in depicting the expression of transcription factors in the developing heart?"

Question three asked, "*How helpful do you believe 3D animations are compared*

to 2D drawings in understanding heart development?" Seven people gave a score of 5,

and four gave a score of 4, indicating the importance of 3D animation in effectively

communicating heart development (Table 3).
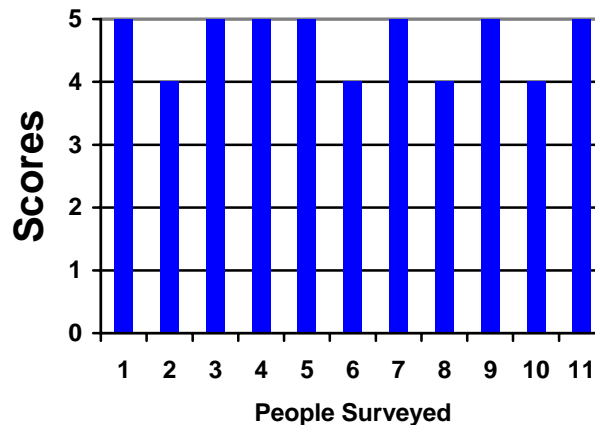
Table 3 – Question three - "How helpful do you believe 3D animations are compared to 2D drawings in understanding heart development?"

The forth question asked, "*How important to you believe it is to have an animation that can be easily updated to include new research?*" Ten of the eleven people surveyed gave a score of 5 for this question, while one gave a score of 4. All of the researchers surveyed appreciated the importance of having an adaptable animation (Table 4).
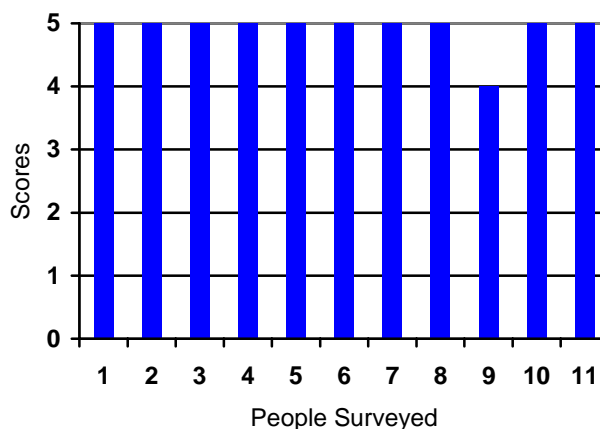
Table 4 – Question 4 - "How important to you believe it is to have an animation that can be easily updated to include new research?"

Question five asked, "*How important do you believe it is to have an animation that can be used for multiple topics?*" Four researchers gave a score of 5, four gave a score of 4, and three gave a score of 3 (Table 5).
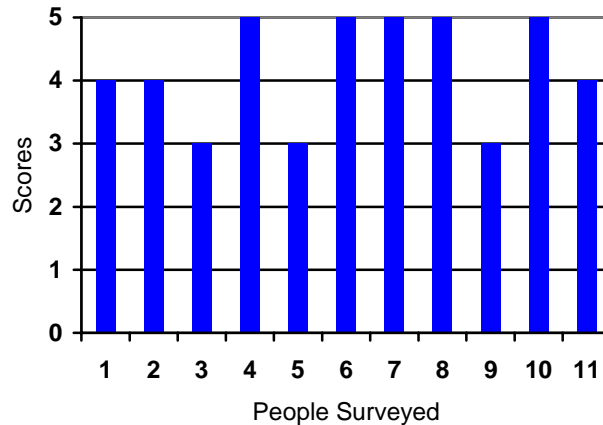
Table 5 – Question five - "How important do you believe it is to have an animation that can be used for multiple topics?"

The sixth question asked, "*How effective was the morphing sequence in showing how the heart develops from the cardiac crescent to the adult heart?*" Four of the people surveyed gave a score of 5, five gave a score of 4, and one gave a score of 3. One person did not answer the question, indicating that he was unsure what was meant by the "morphing sequence" (Table 6).
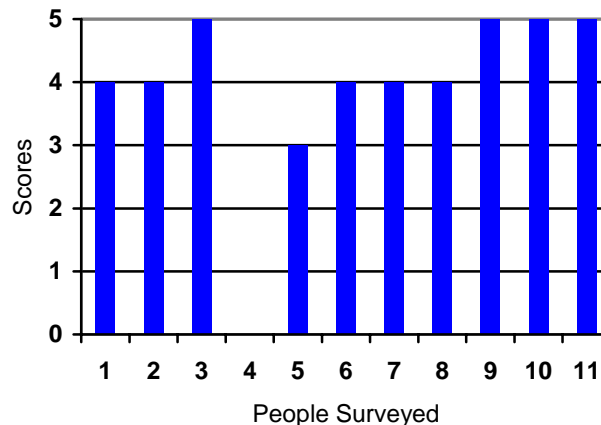
Table 6 – Question six - "How effective was the morphing sequence in showing how the heart develops from the cardiac crescent to the adult heart?"

Question seven asked, *"How effective were the colors chosen to highlight specific areas of the developing heart?"* Two people gave a score of 5, one gave a score of 4, six gave a score of 3, one gave a score of 2, and one gave a score of 1. The general consensus was some of the colors were too subtle, and needed to be more intense (Table 7).
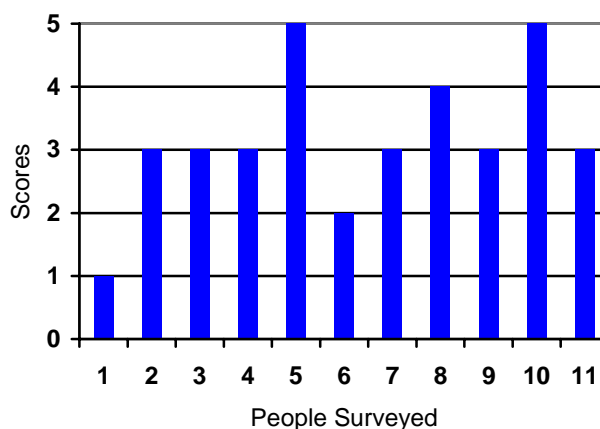
Table 7 – Question seven- "How effective were the colors chosen to highlight specific areas of the developing heart?"

The eighth question asked, "*How effective were the labels in the animations?*" One person surveyed gave a score of 5, four gave a score of 4, five gave a score of 3, and one gave a score of 2 (Table 8).
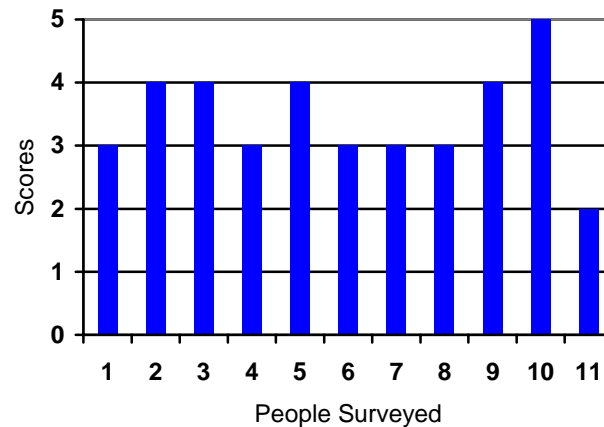
Table 8 – Question eight - "How effective were the labels in the animations?"

Question nine asked, "*How appropriate was the content of the animations for an audience knowledgeable about cardiac morphology?" (An answer of 5 would indicate that the animations were neither too simplistic nor too advanced for the audience.)*" Five researched gave a score of 5, four gave a score of 4, one gave a score of 3, and one gave a score of 2 (Table 9).
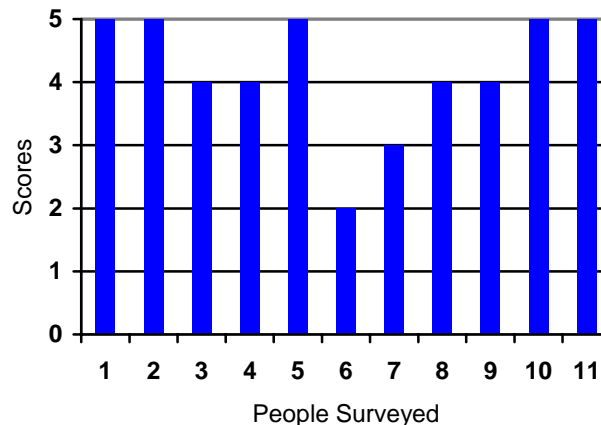
Table 9 – Question nine - "How appropriate was the content of the animations for an audience knowledgeable about cardiac morphology?"

The last question asked, *"How effective were the durations of the animations?"* Two people gave a score of 5, six gave a score of 4, one gave a score of 3, and one gave a score of 2. One of the people surveyed did not answer the question, indicating that the appropriate length of the video would depend on the context for which it was being used (Table 10).
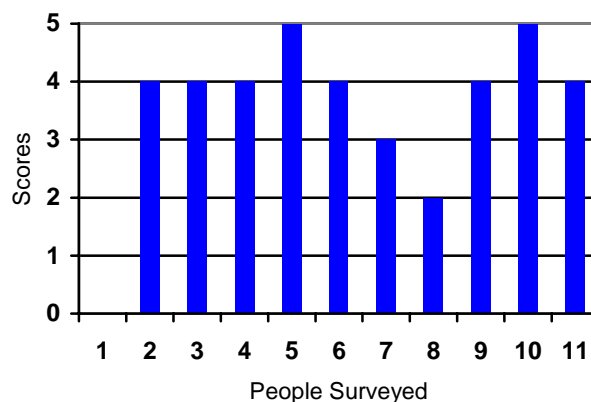
Table 10 – Question ten - "How effective were the durations of the animations?"

Overall, the survey results were highly positive (Table 11). Some critiques included suggestions to label the embryonic day and alter the order of transcription factor labels. While valid points, these ideas were discussed before the animation was created, and decided against. The colors chosen were thought to be too subtle, especially in the transcription factor video. In all, the researchers surveyed appreciated the idea that animations could be easily updated. They also stressed the importance of 3D animation in depicting heart development.
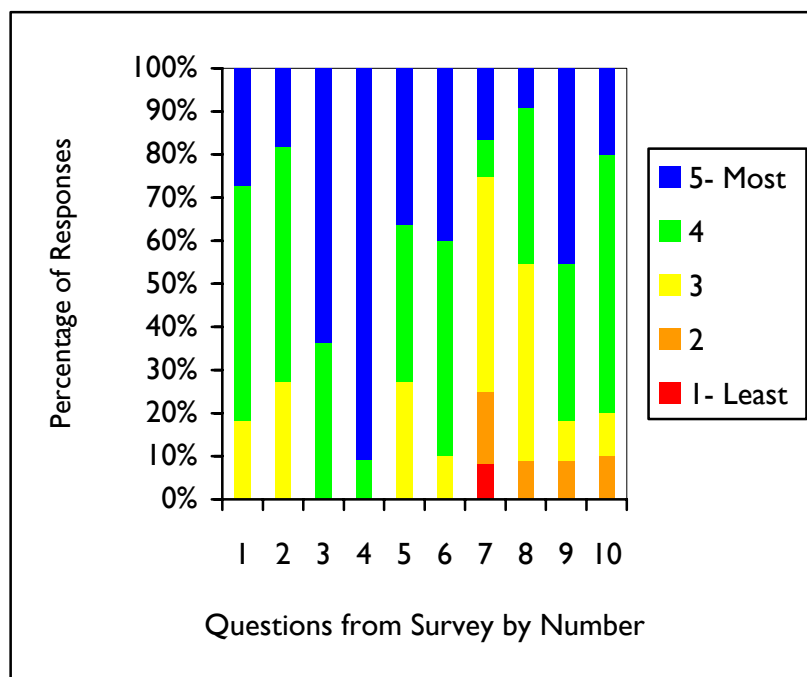
Table 11 – Combined survey results.

CHAPTER FIVE
Conclusions and Recommendations

<u>CONCLUSION</u>

This thesis endeavored to develop a technique for easily altering 3D animations using 2D methods, enabling researchers to use the same model to visualize multiple topics. This process also eliminates the need to go back into a 3D software program to make changes or update an animation.

In order to meet my first objective, I researched possible subject matters with the Olson Laboratory to determine what two topics would be the basis of the animations. Second, I created illustrations and three-dimensional models that accurately represent the anatomical stages of heart development. Finally, using 3ds max®, I created a generic three-dimensional animation of heart development, which was enhanced using two-dimensional techniques to produce two distinct videos.

The first video depicts cardiac morphogenesis with color-coding of morphologically related regions, achieved in Adobe After Effects®. The second video incorporates the same 3D animation, but shows the relationship of crucial transcription factors to proper heart development. Again, After Effects® allowed two-dimensional components to redirect the purpose of the animation.

The completed animations were edited and finalized using Adobe Premiere®, and placed on CD. After completion, a group of molecular biologists and cardiologists viewed the final videos and answered a ten question survey. The results were highly positive, with those surveyed agreeing that three-dimensional animations are preferred to

illustration in explaining the process of heart development. The group also embraced

the idea of easily altering an animation to reflect the fast pace of research today.

SUGGESTIONS FOR FURTHER RESEARCH

Cardiovascular disease and CHD are the leading causes of death in both adults

and children in the United States, opening up many avenues for future study in the field

of heart research. These videos explain the process of heart development, and the

transcription factors involved, however, many other concepts could be presented. By

using the 3D models available, one could easily test this thesis by creating a new

animation entirely in After Effects®.

Dr. Olson expressed a desire for a third animation based on the Transcription

Factor Video already created. To take the video one step further, he suggested indicating

what heart defect would present itself if a mutation or deletion occurred in one of the

factors. One could easily accomplish this by inserting 2D illustrations of heart defects

into the existing animation.

Yet another idea for future research is to expand on the idea of incorporating 2D

effects into 3D animations. Whatever the subject matter, creating basic 3D models and

leaving the complicated surface mapping and animation effects to a 2D editing program,

can expedite and simplify the entire process. This process saves both the animator and

client from the cumbersome and costly practices typically associated with developing a

video.

APPENDIX A
Storyboards

Project: Heart Animation

Heart Development (E7.5 - Birth)

Main Anim. #1   Page: 1

Panel 1: Heart Development (Title)
Title
Keep simple → Dr. Olson will want to go directly into animation.

Panel 2: E7.5
Fade in. Go through one set of morphing. With color?

Panel 3: pinch ←

Panel 4: elongate ↑↓
E8

Panel 5: AVC RV LV moves back
arteries to side
E8.5 - E9.5

Panel 6: Atria wrap forward.
E10 - E12
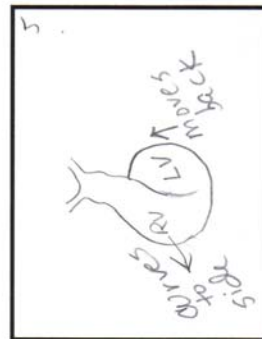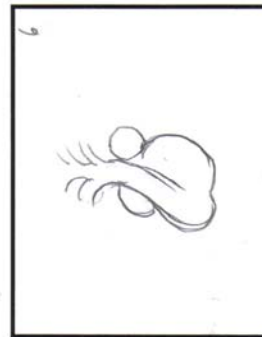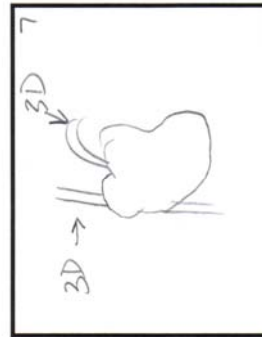
Panel 7: 3D → ← 3D
Vena cava + aorta shown in 3D growing out.
Use 2D Adult heart.
Birth

Panel 8: E7.5 Cardiac crescent
Cardiac crescent fades in. Labeling.

Ryan Carre, 214-821-8995   Date: 9/8/04

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Project: Heart Animation

9
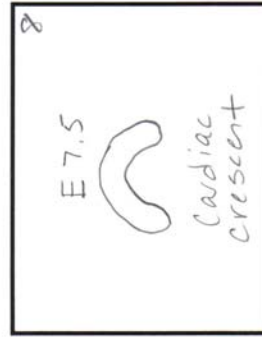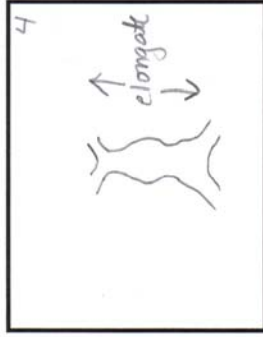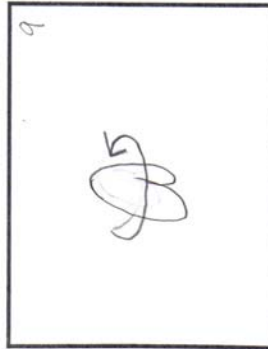
Rotate.
Come to stop in
front.

10
Atria
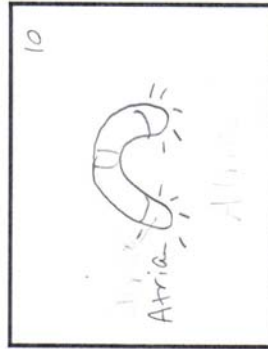
Label.
Either under or
pointing.
Area "glows".
(Have color present →
just ↑ saturation
when labeling)

11
Ventricle

12
Conotruncus

13

E Morph

14
E8
Heart Tube
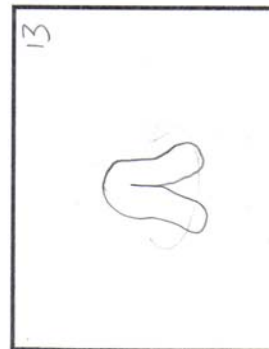
Repeat steps
8 – 13 with
next three
stages.

15
Repeat
w/ Heart +
Looping
+ Formation
Chamber + septation

16

morphing from
stage E10 to
Birth

Open models
to label
inner anatomy.

E12 - Birth 17

Adult Heart

by
Ryan Carre
© 2004

Credits
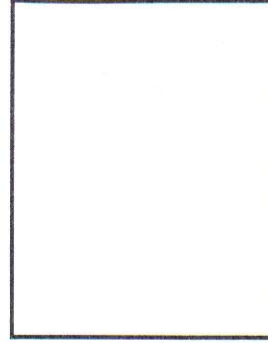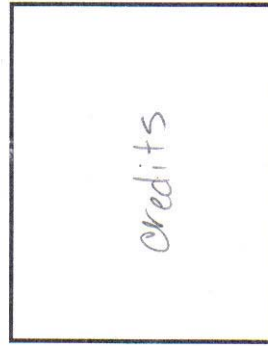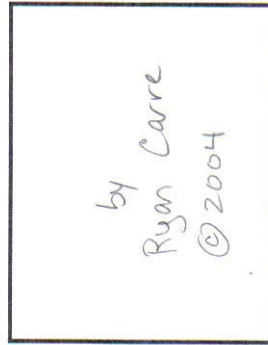
Z.D.

Go through
labeling →
no rotation.
Fade out.
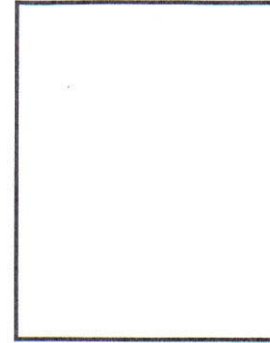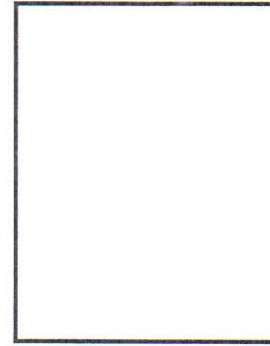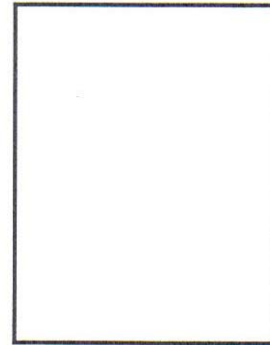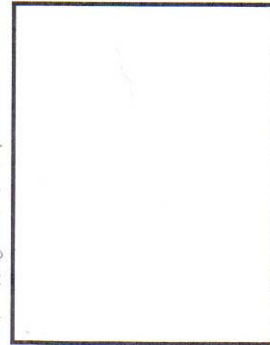
Project: Heart Animation

Expression Patterns of "Factors"

Main Anim. #2   Page: 1

"Expression of Trans. Factors in Heart Develop"

Title. Fades in/out.

HAND 2

Subtitle

Morph through all stages.

HAND 1
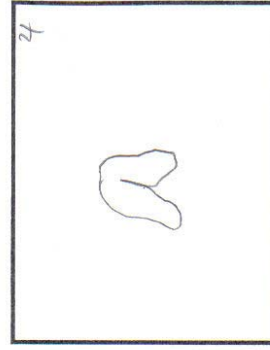
Subtitle
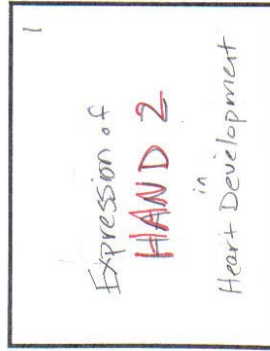
Show locations of individual factors.

morph...show all stages.

Continue for other factors.

* Should I label anatomy in these? How?

Ryan Carre, 214-821-8995   Date:

Project: Heart Animation   HAND 2 — SUB-ANIMATION   Page: 1
- Can be shown in total or alone.

1
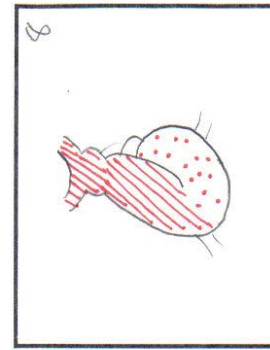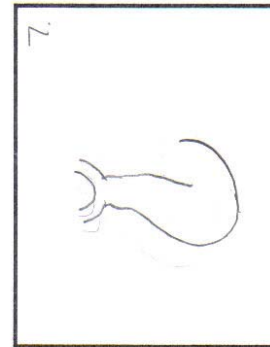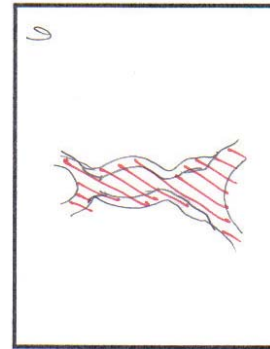Expression of
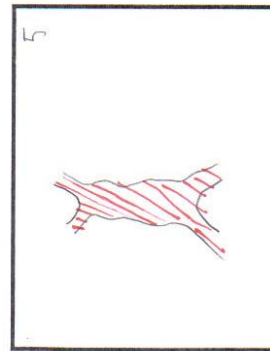HAND 2
in
Heart Development
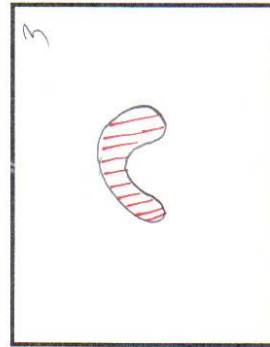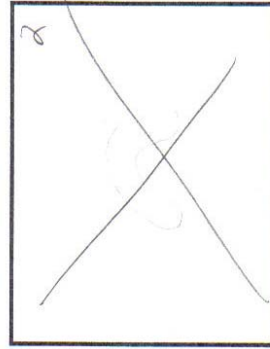
Subtitle

2
Go through
morphing?

3
Fade in.
Local color w/
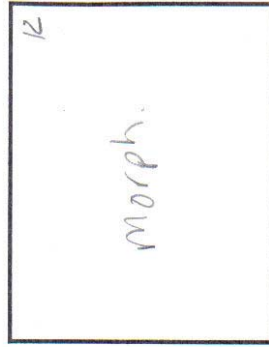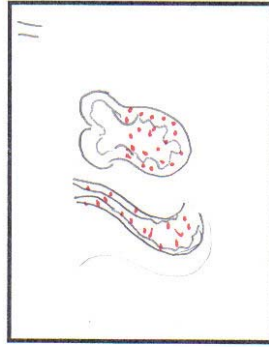highlighted
area of HAND 2.

4
morph

5

6
Open HAND 2
Show inside

Close → morph.

7
morph.

8

Project: Heart Animation

Page: 2

9 morph	10	11	12 morph
13	14	15	16
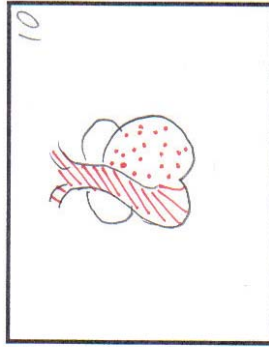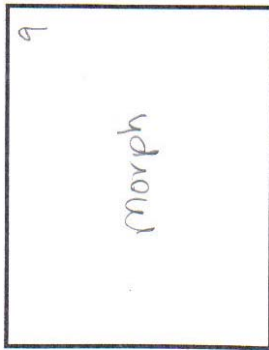
Open.
Not in cardiac cushions
Close.

Ryan Carre, 214-821-8995

Date: 9/8

yellow

Ana Barbo

eHAND — HAND1

44
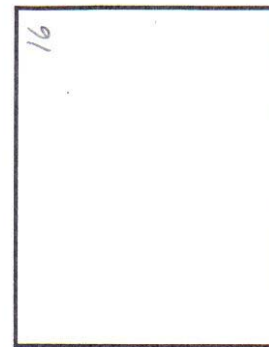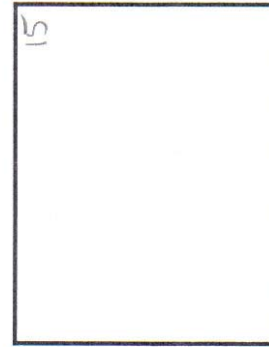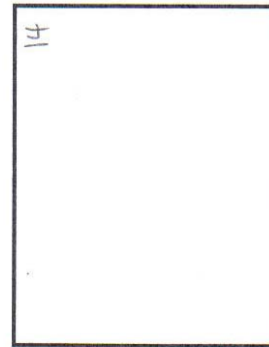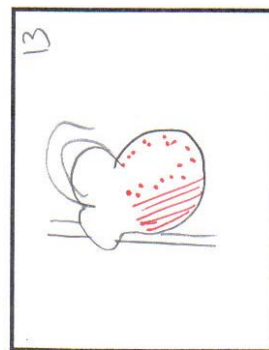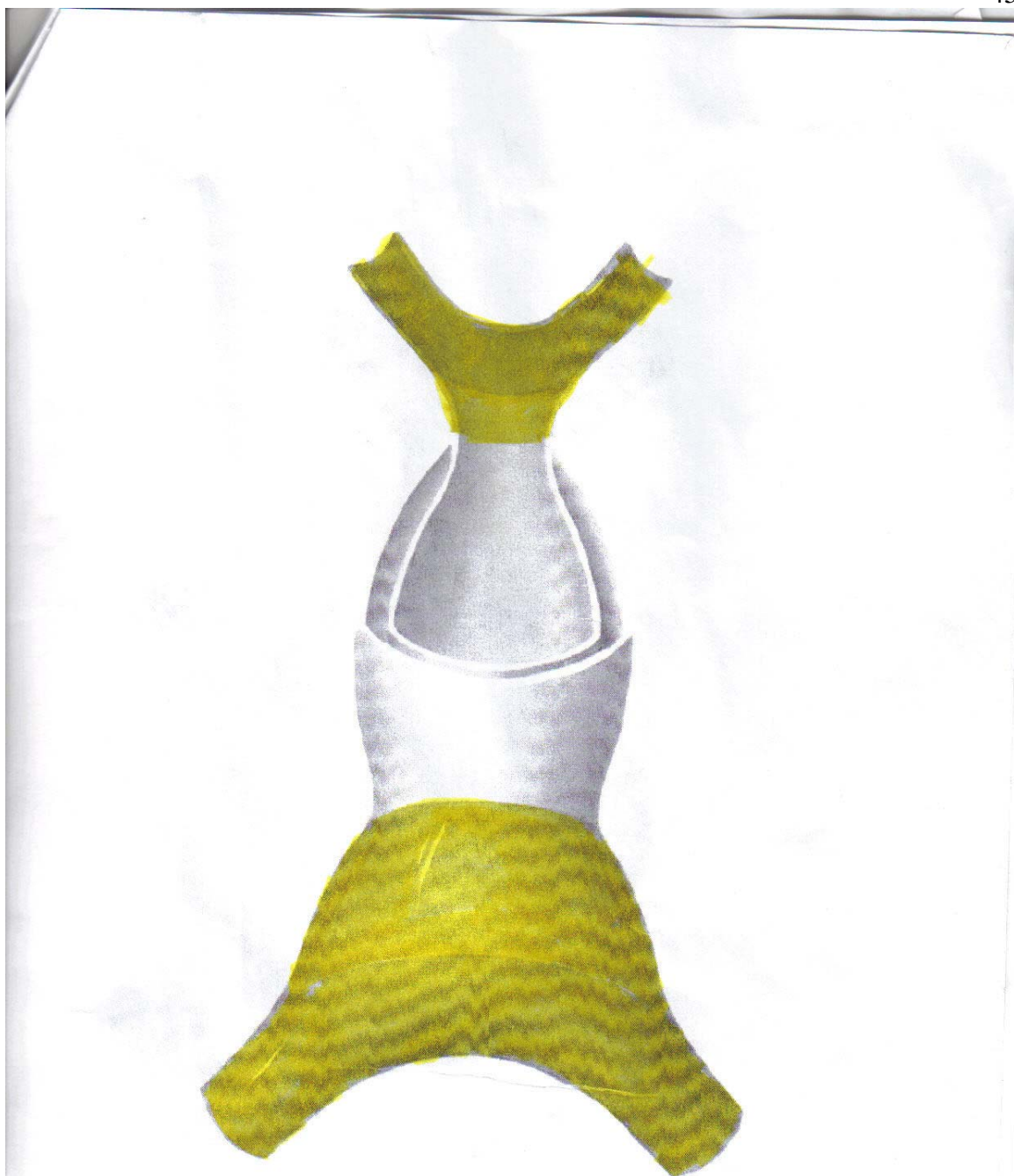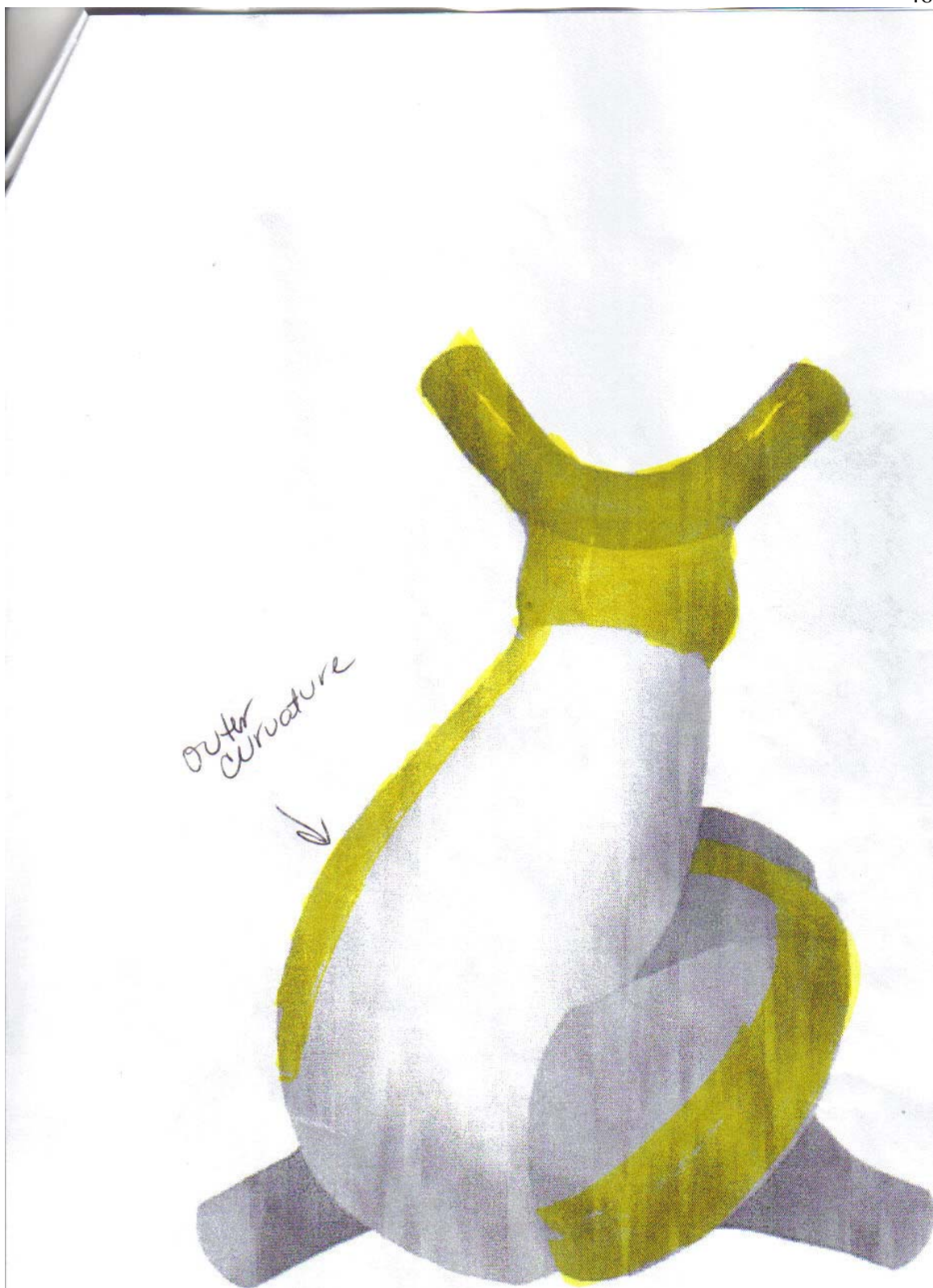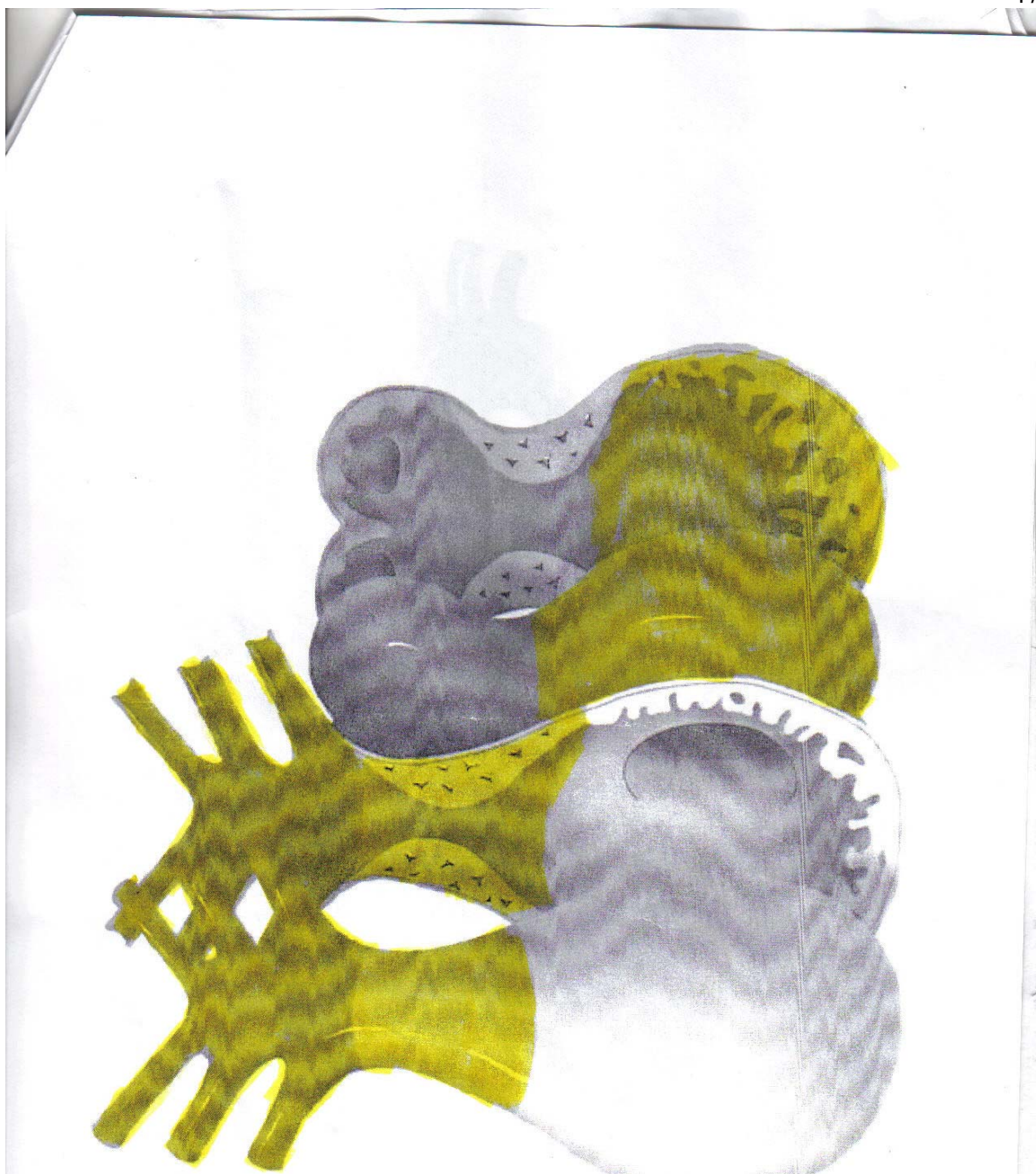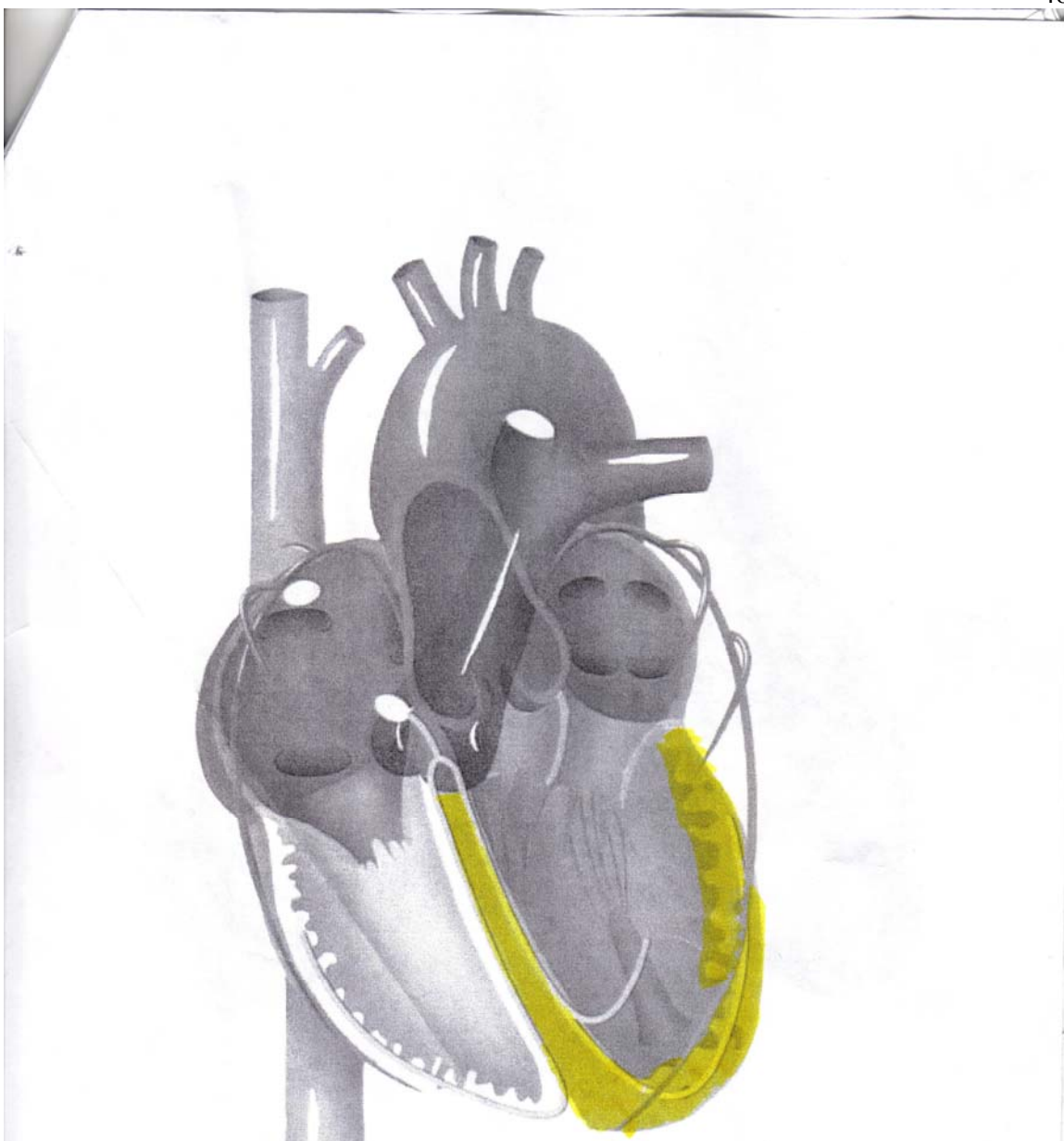
outer
curvature

blue

Ana Barbosa

dHAND = HAND 2
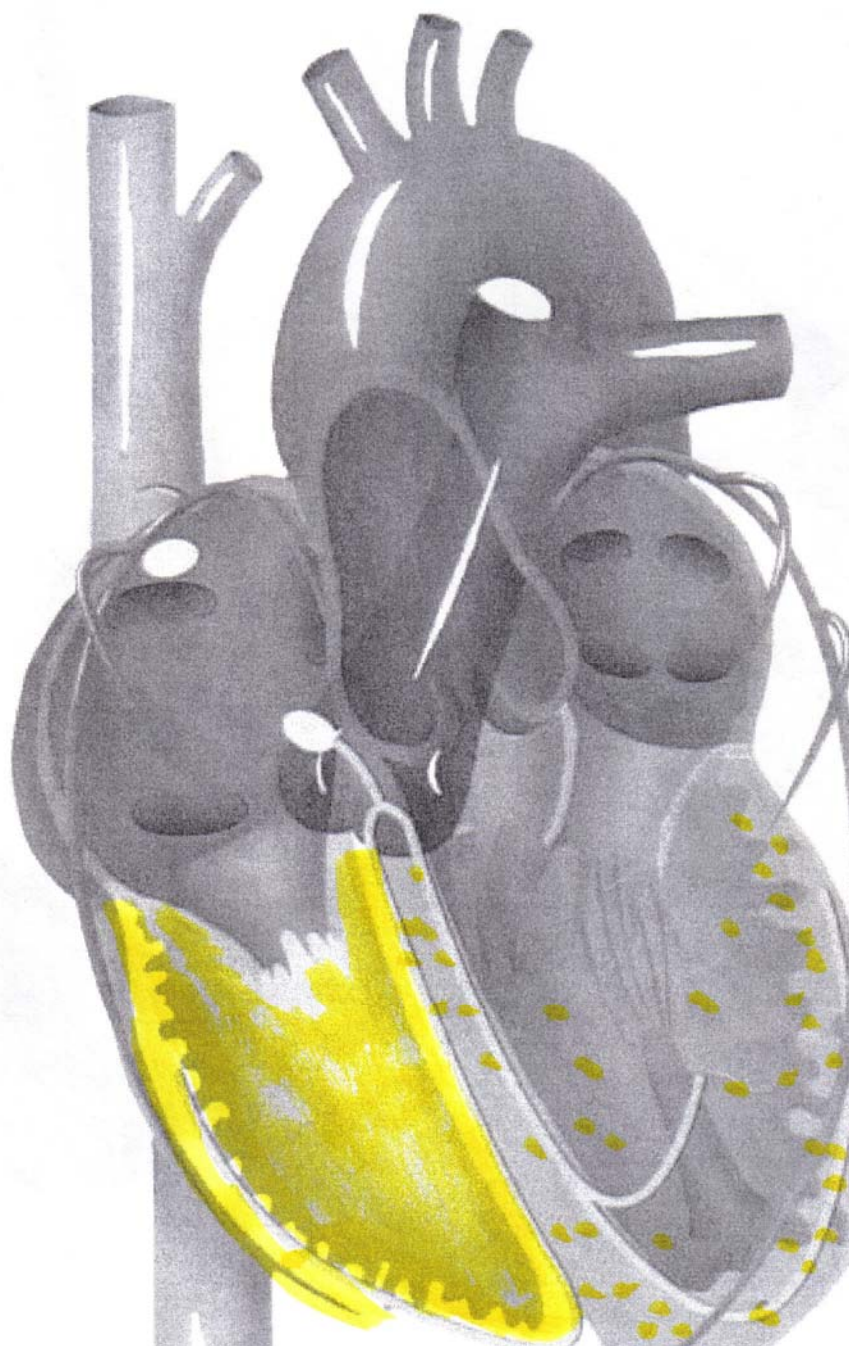
red   Myocardin
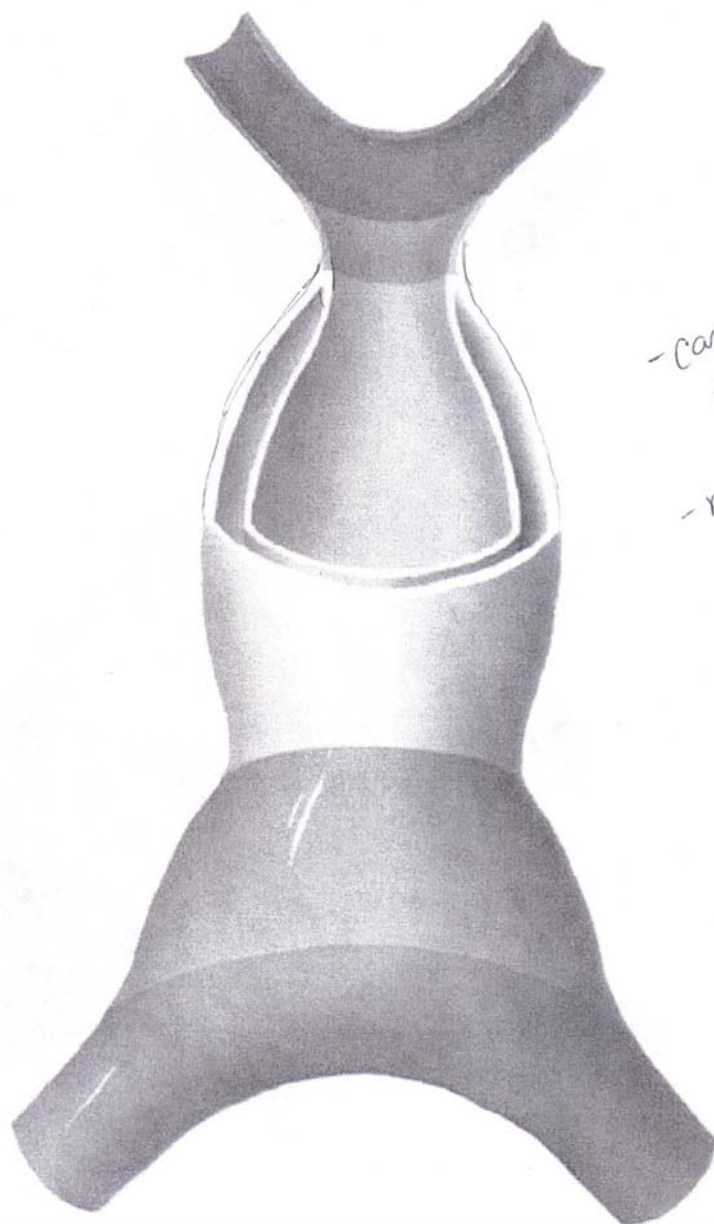
- can detach polygons
- make copy of outer shell make sm

Esther.
Creemer
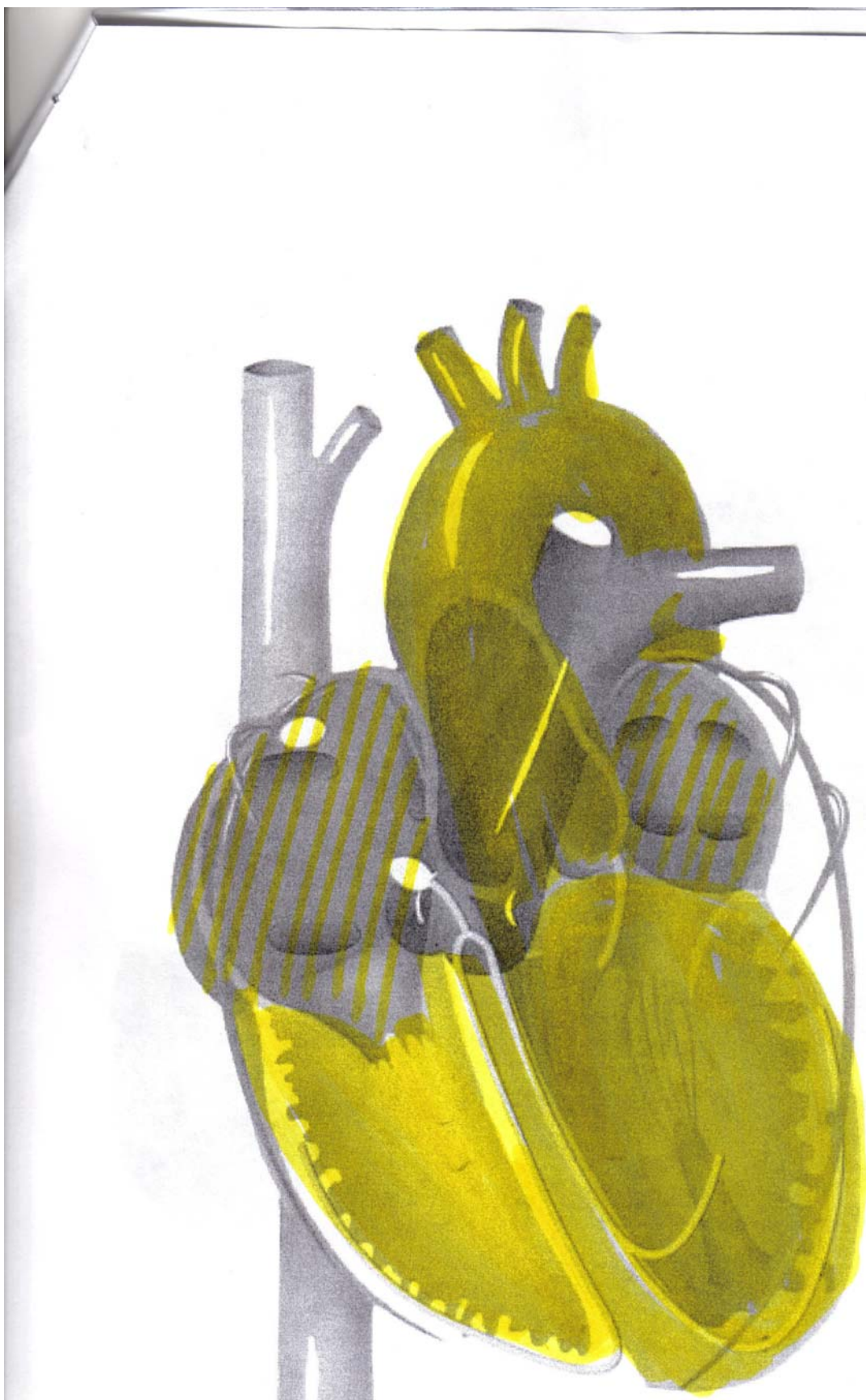
green

NKX2.5
Expression
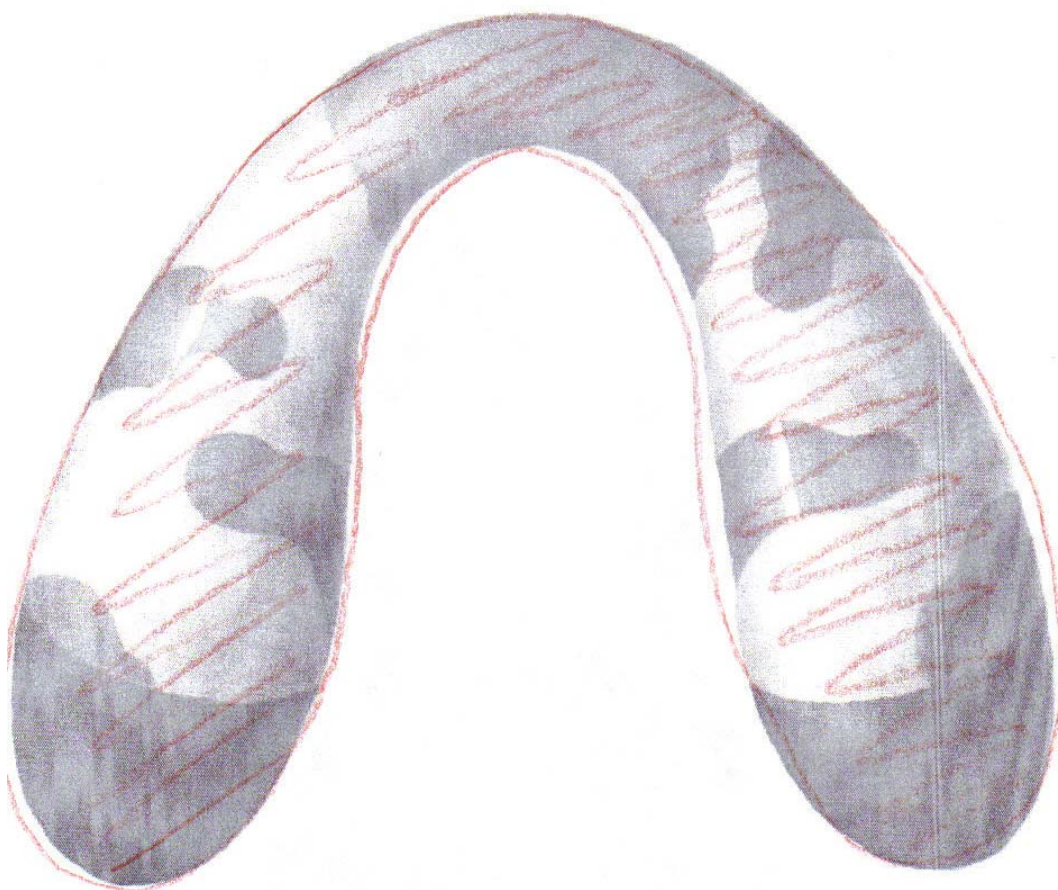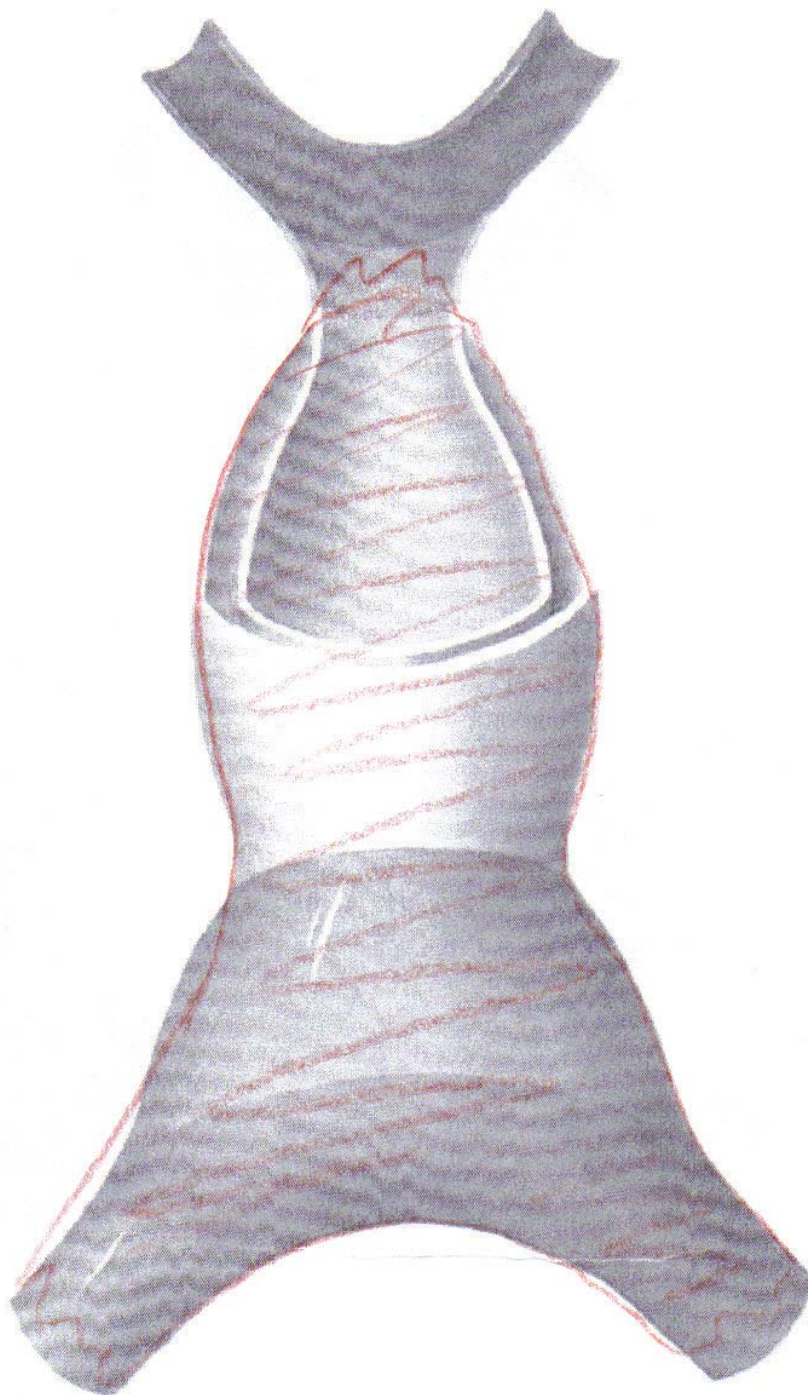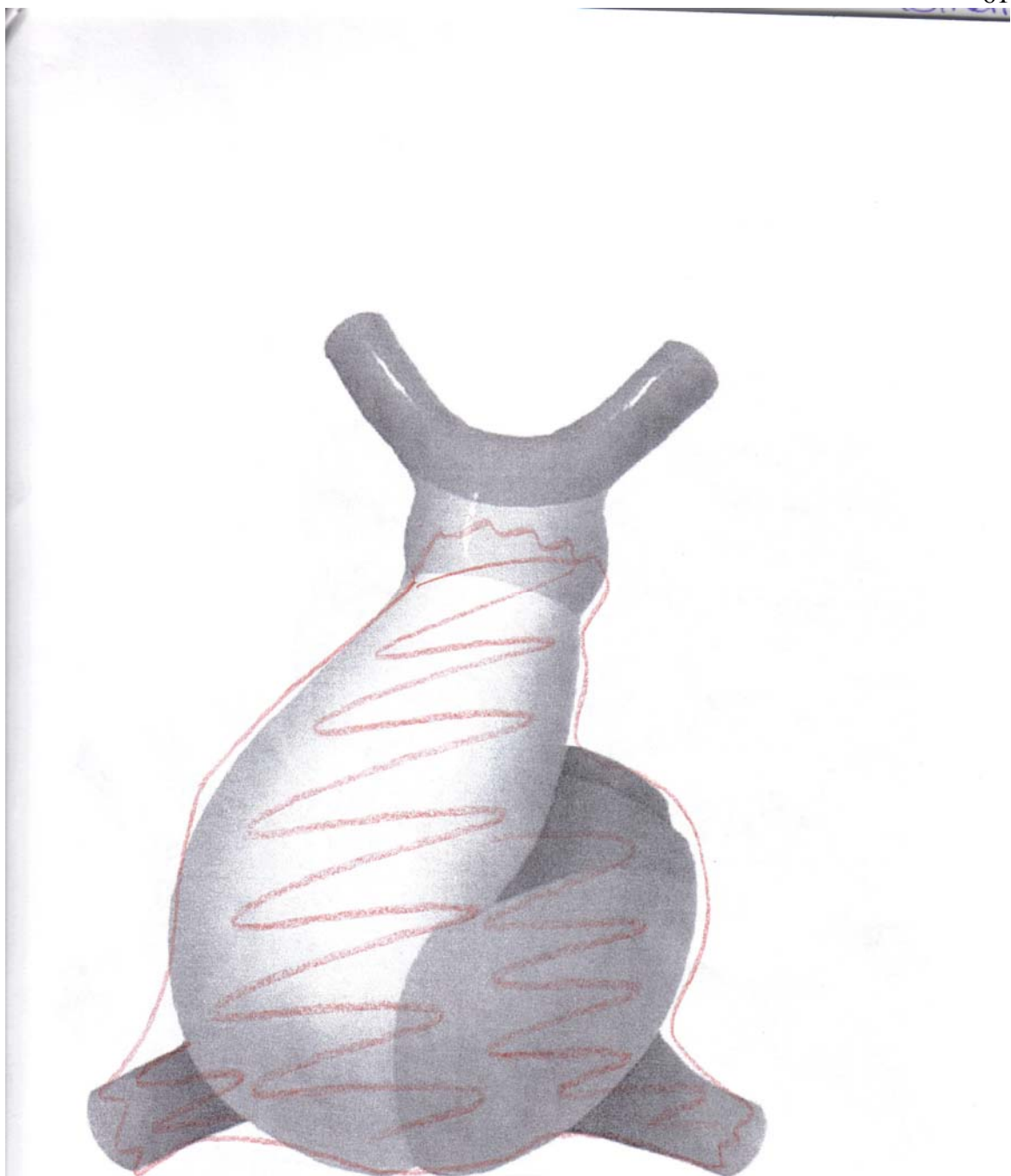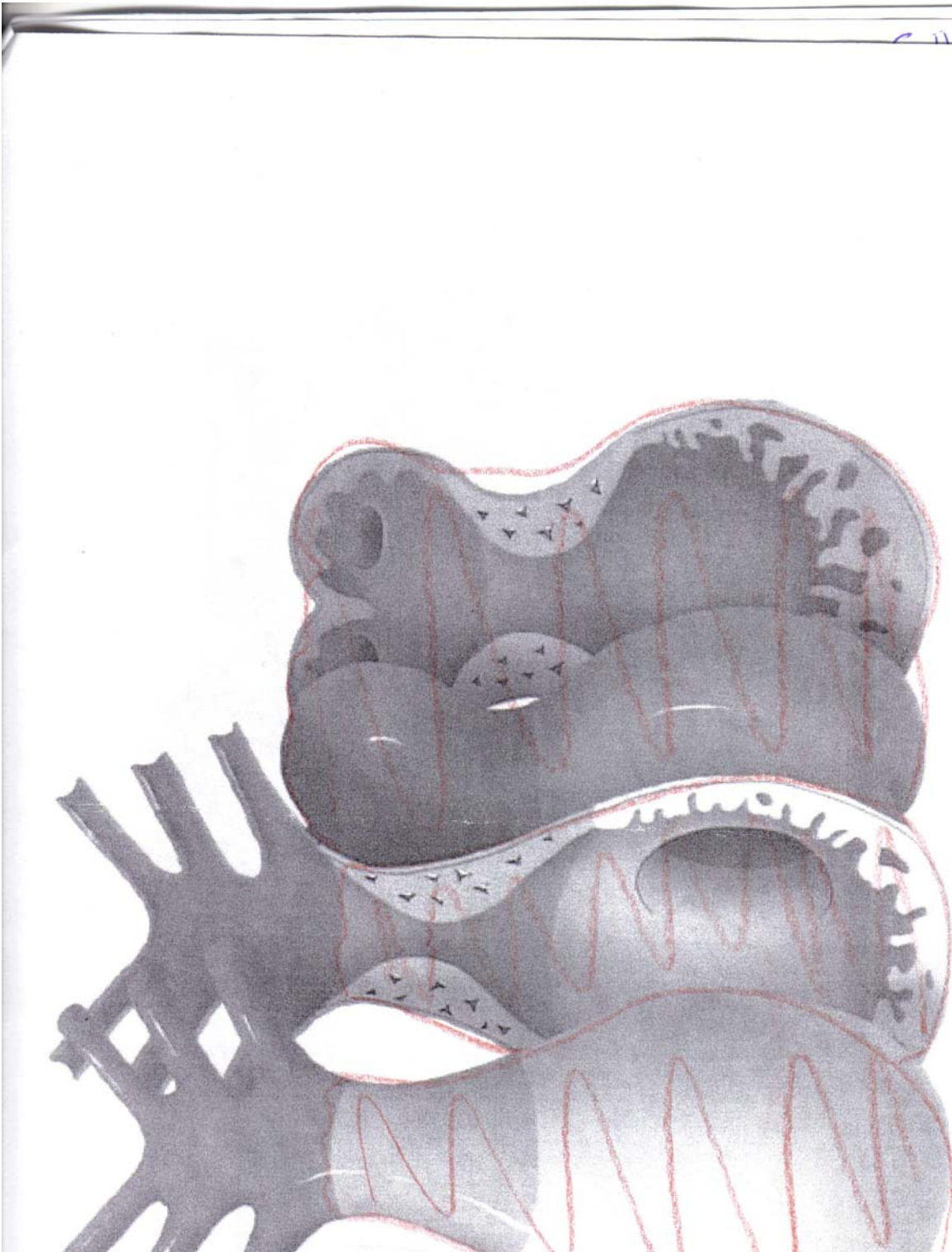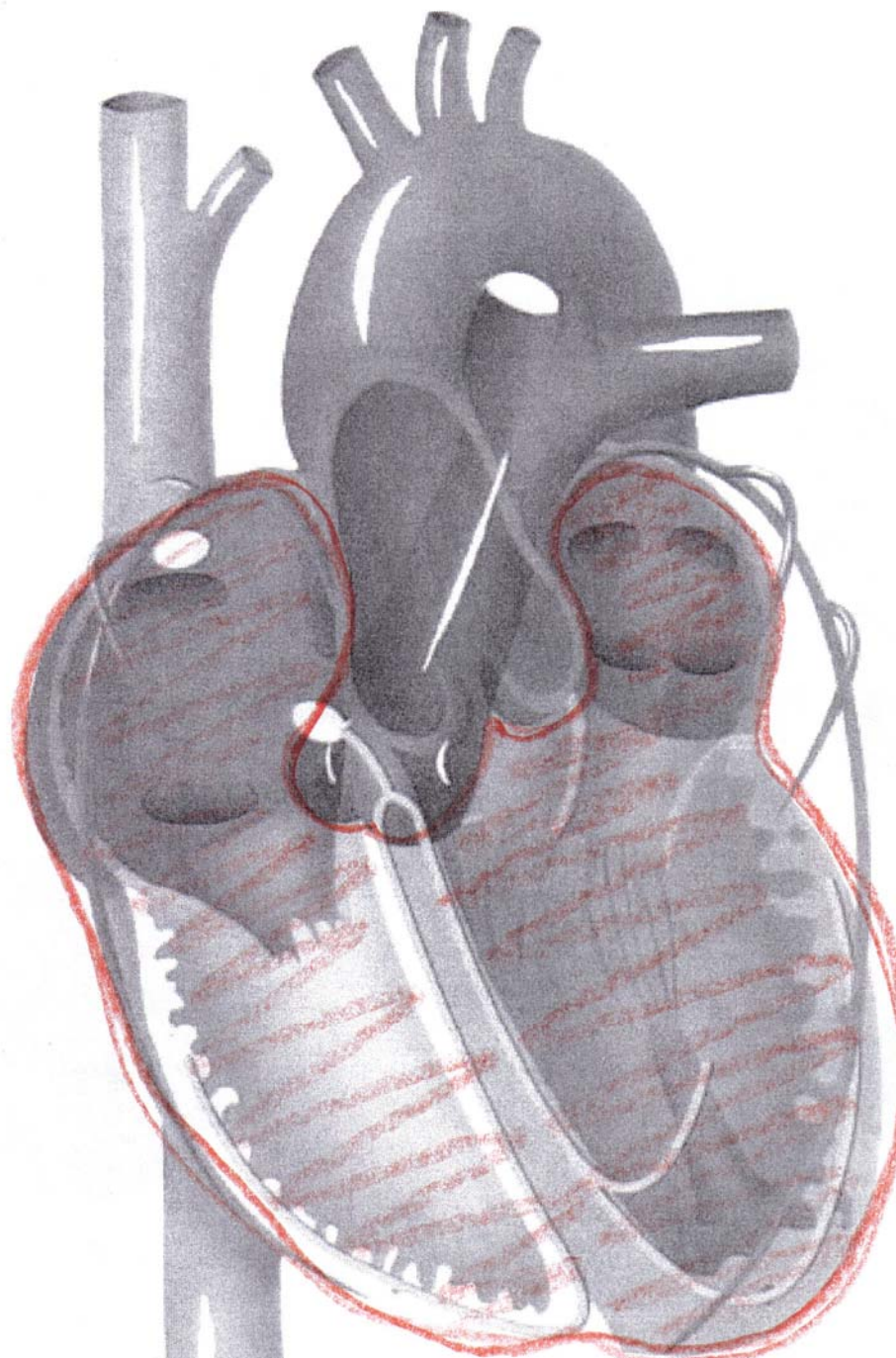
orange
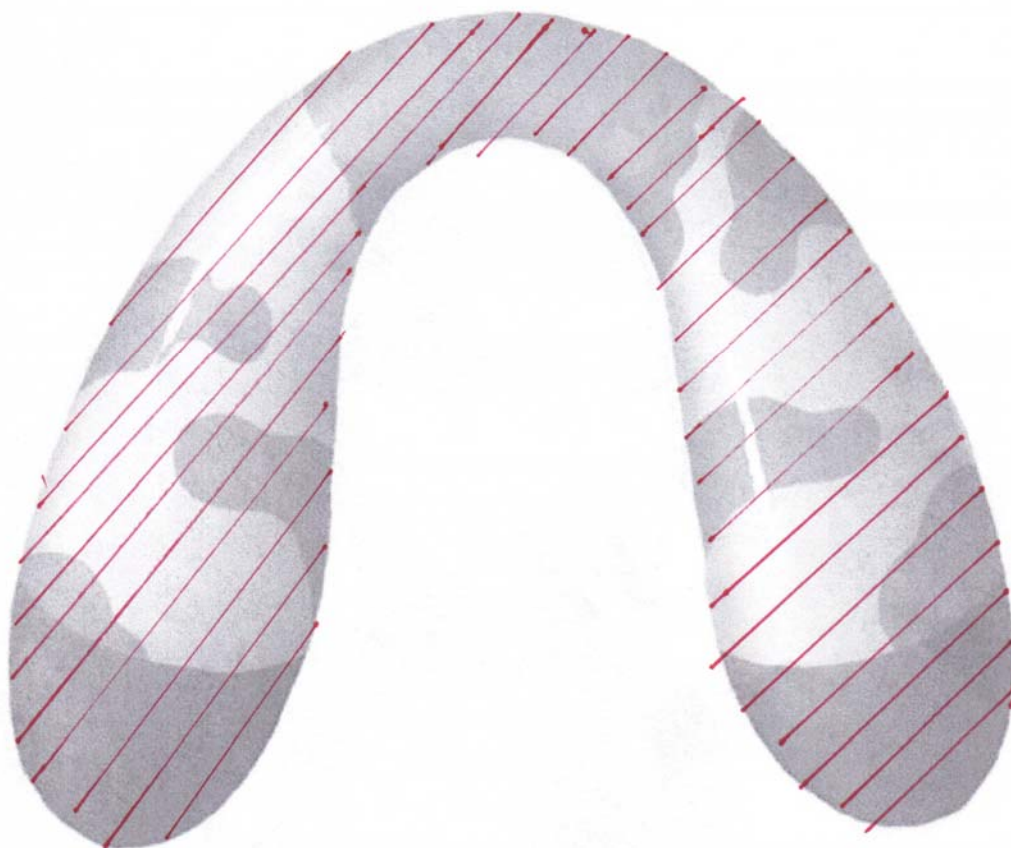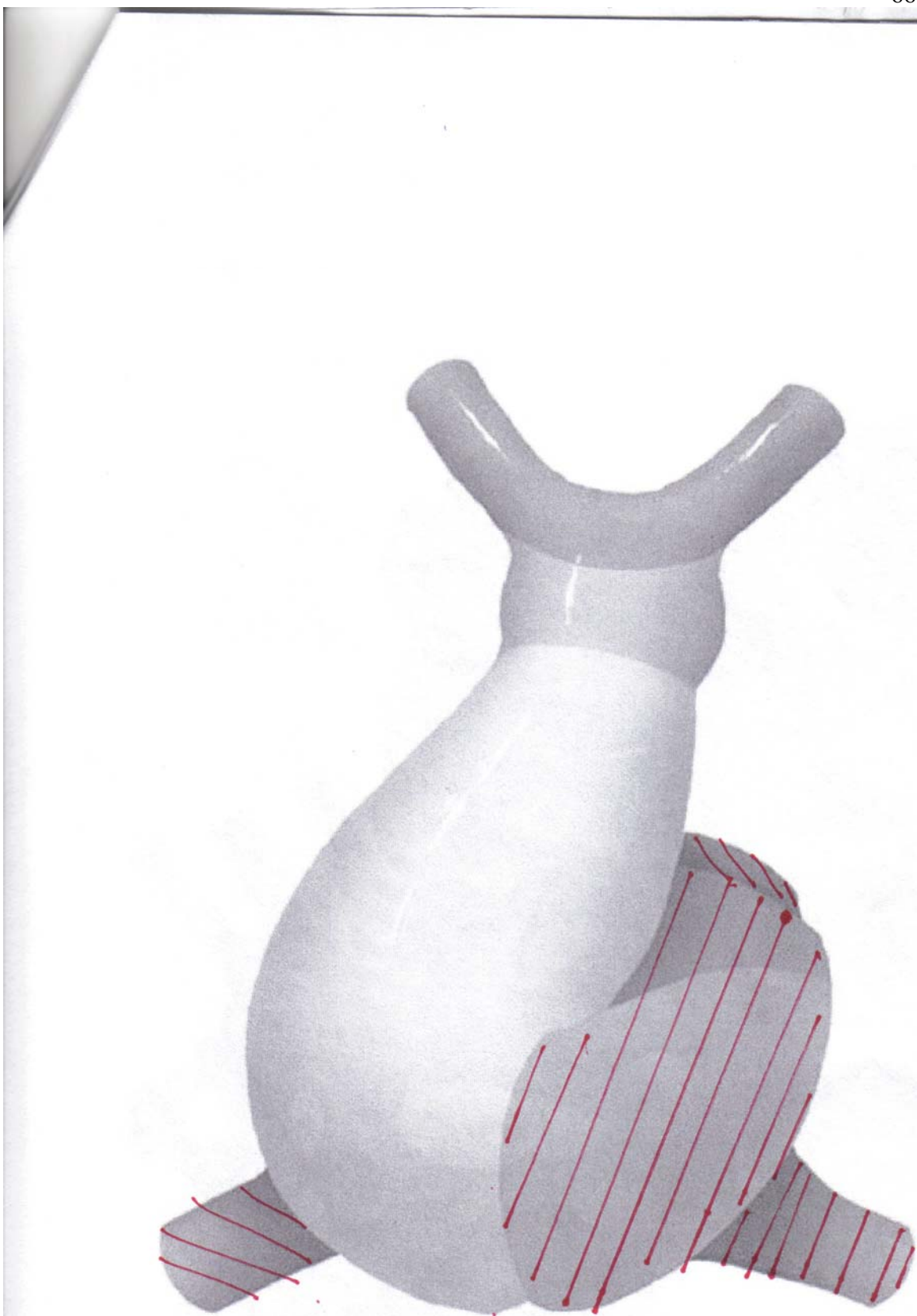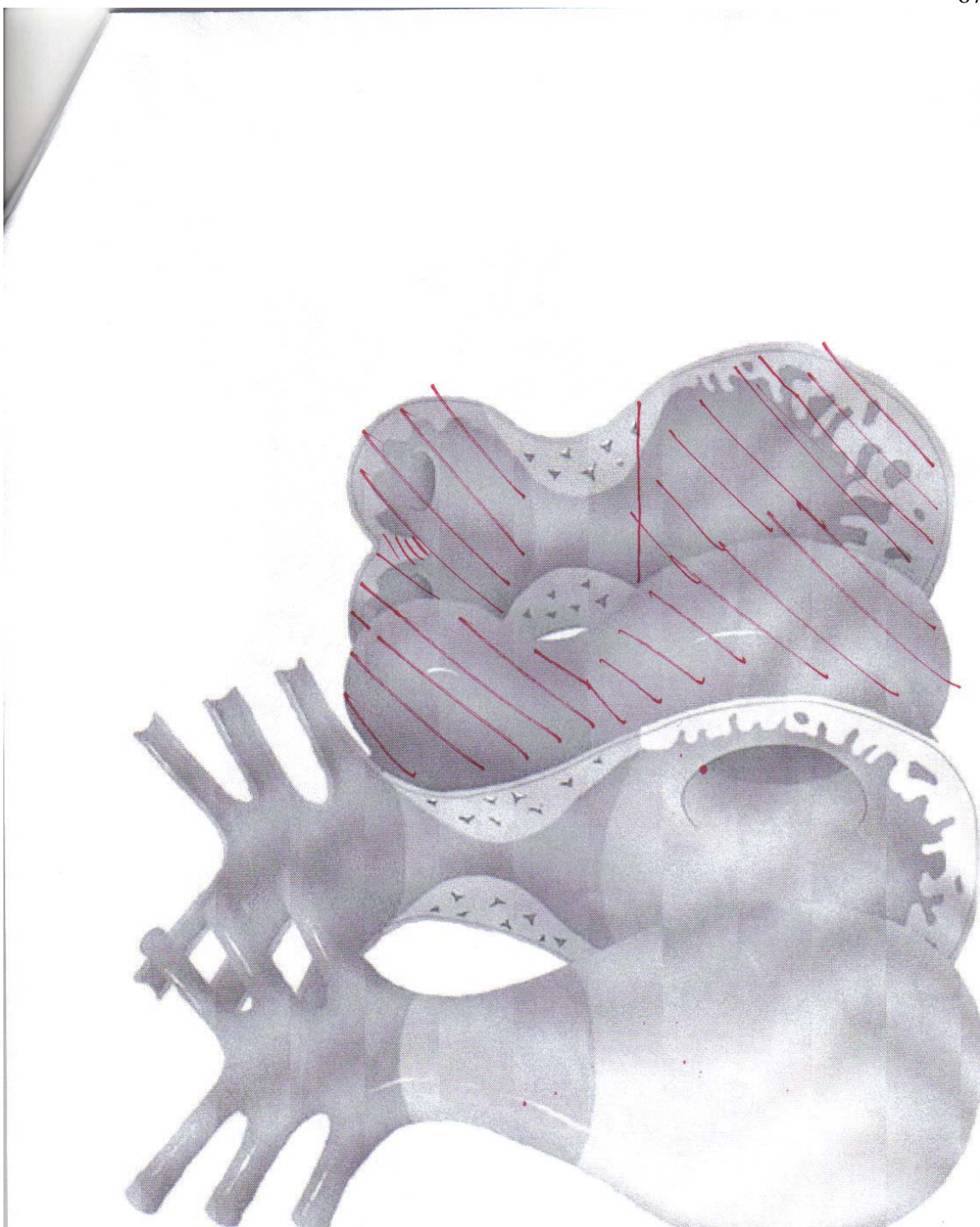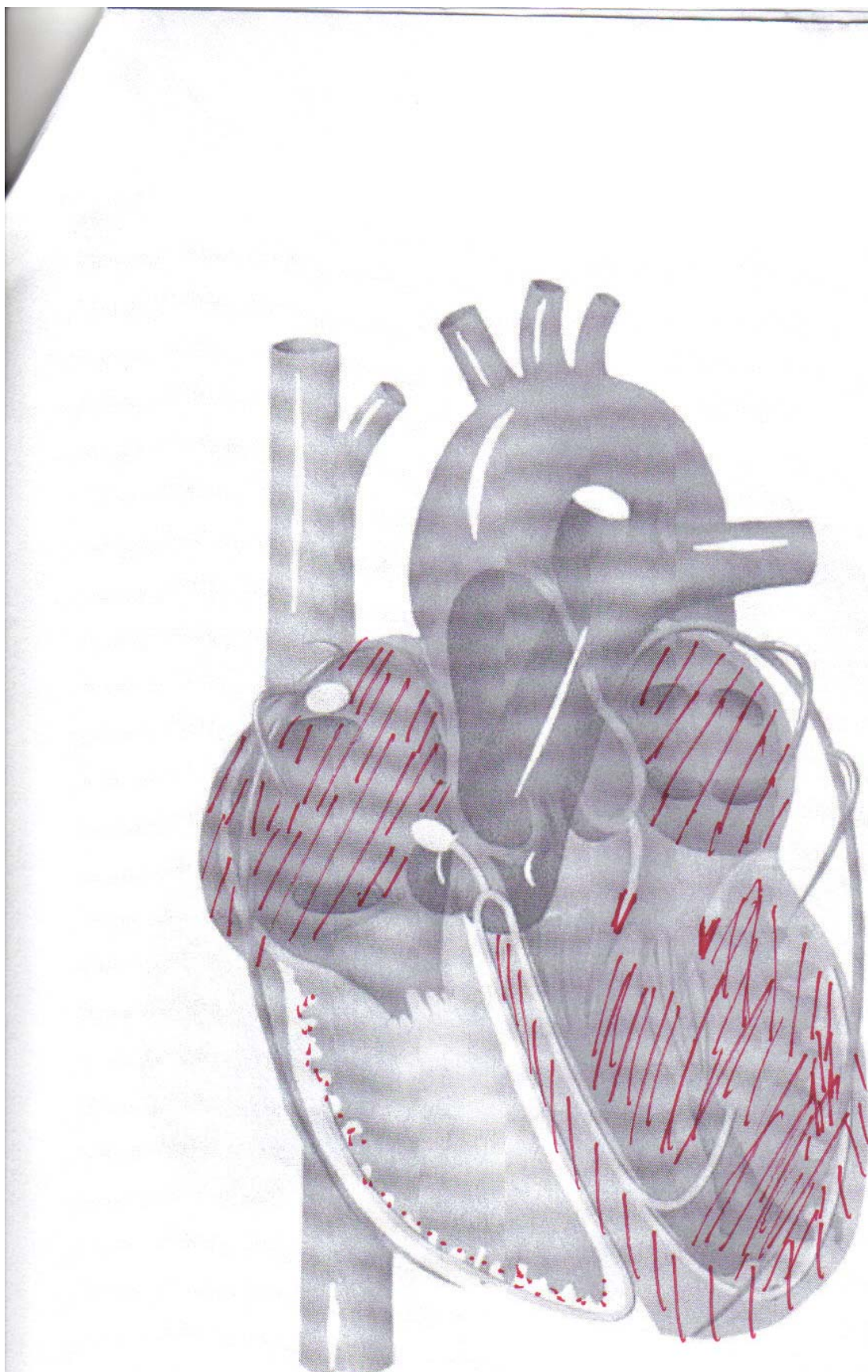
Ana Barbosa

Tbx5

purple

GATA4
Mei Xin

HRTI light blue

Osam
Naka
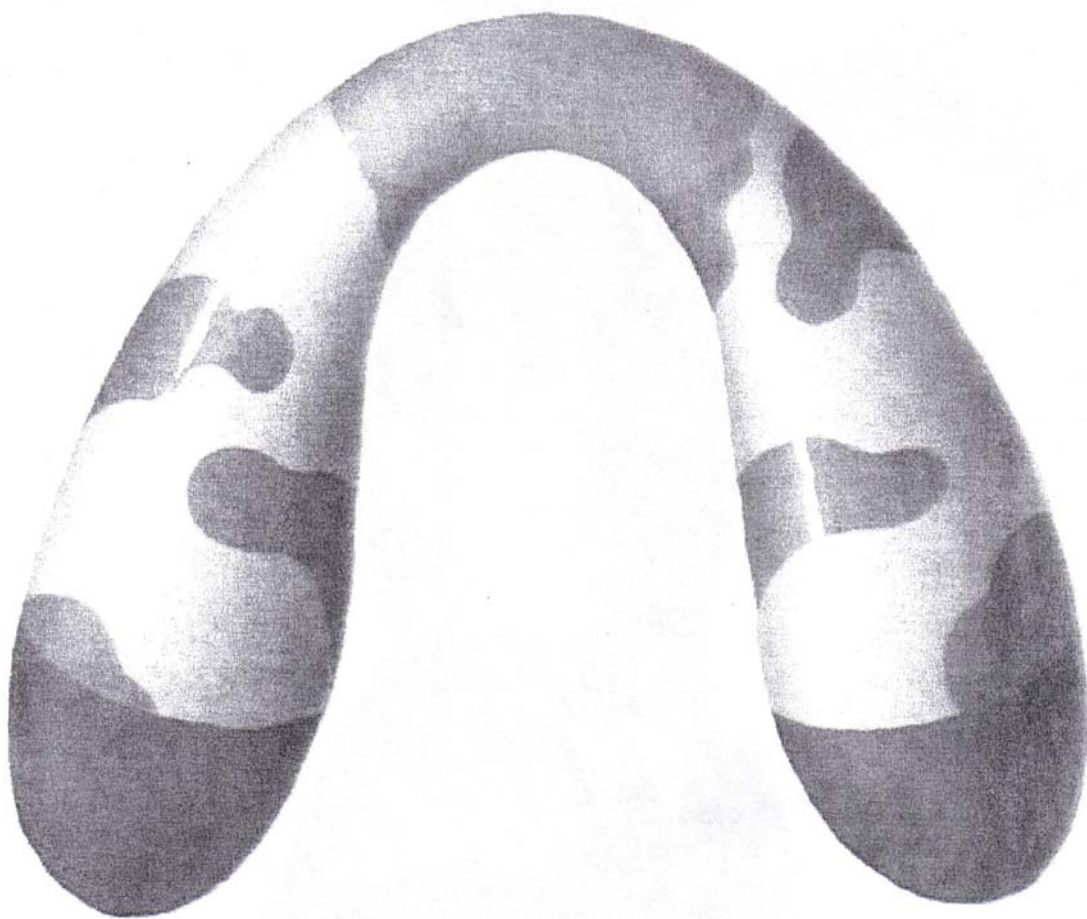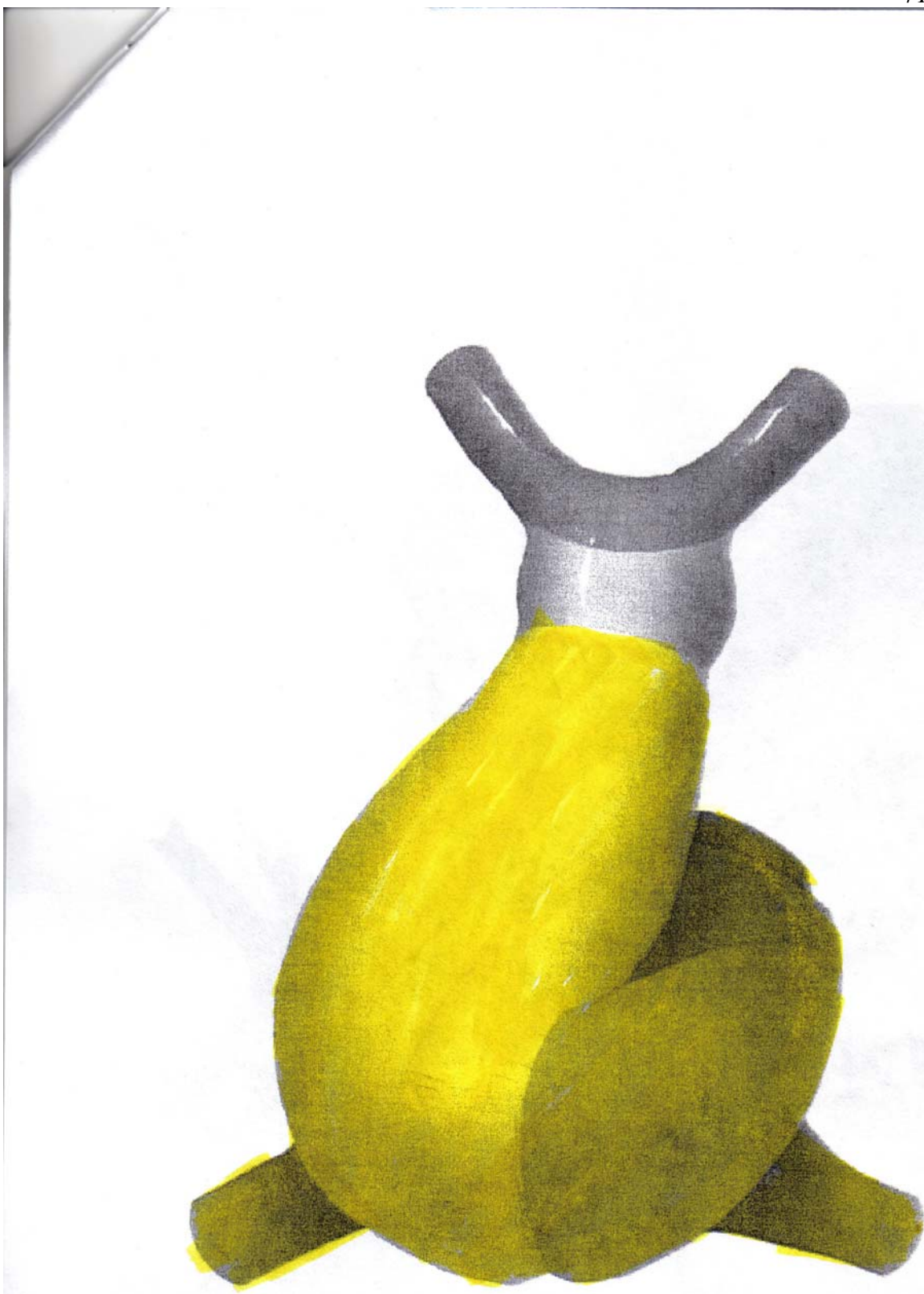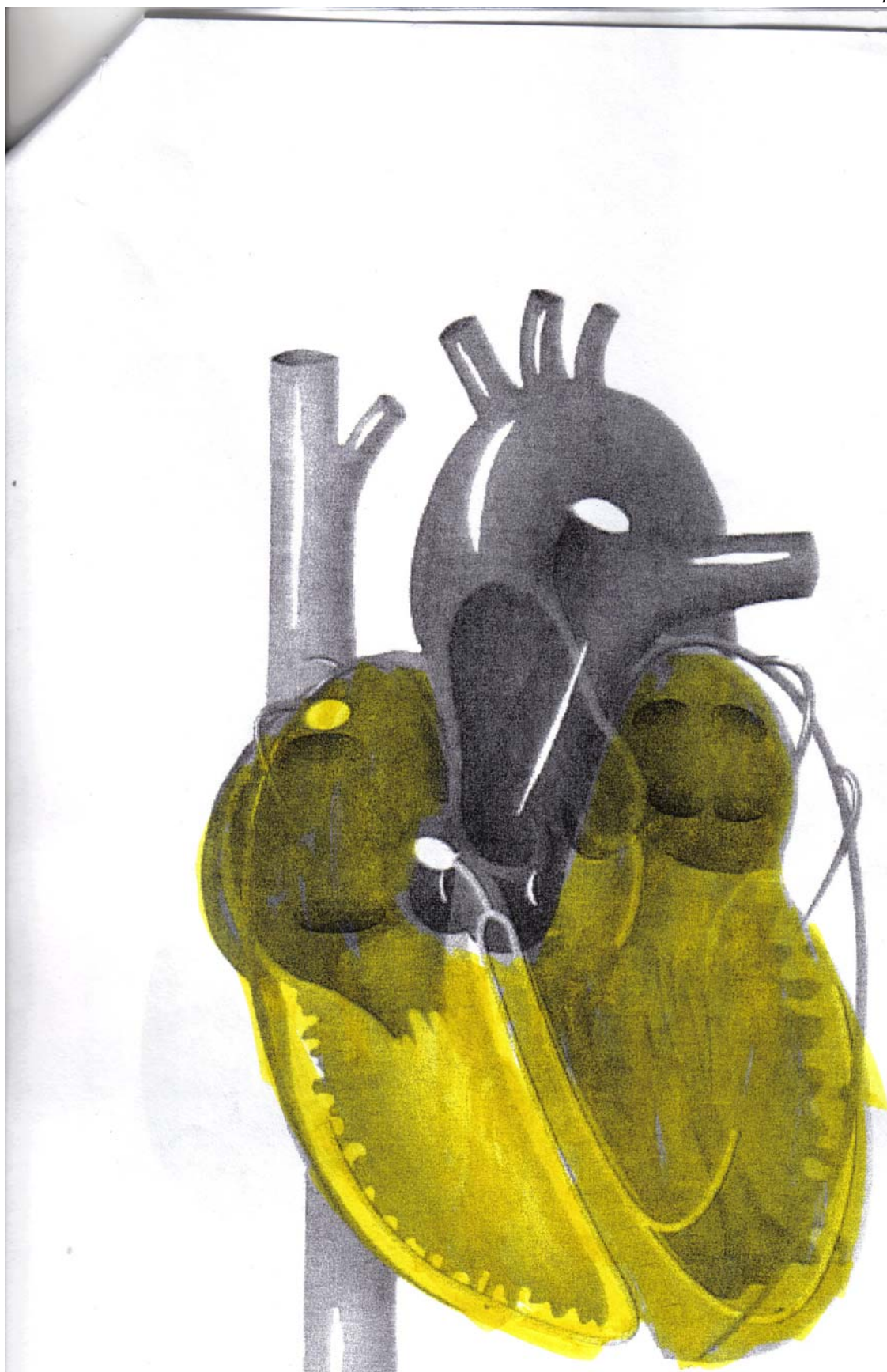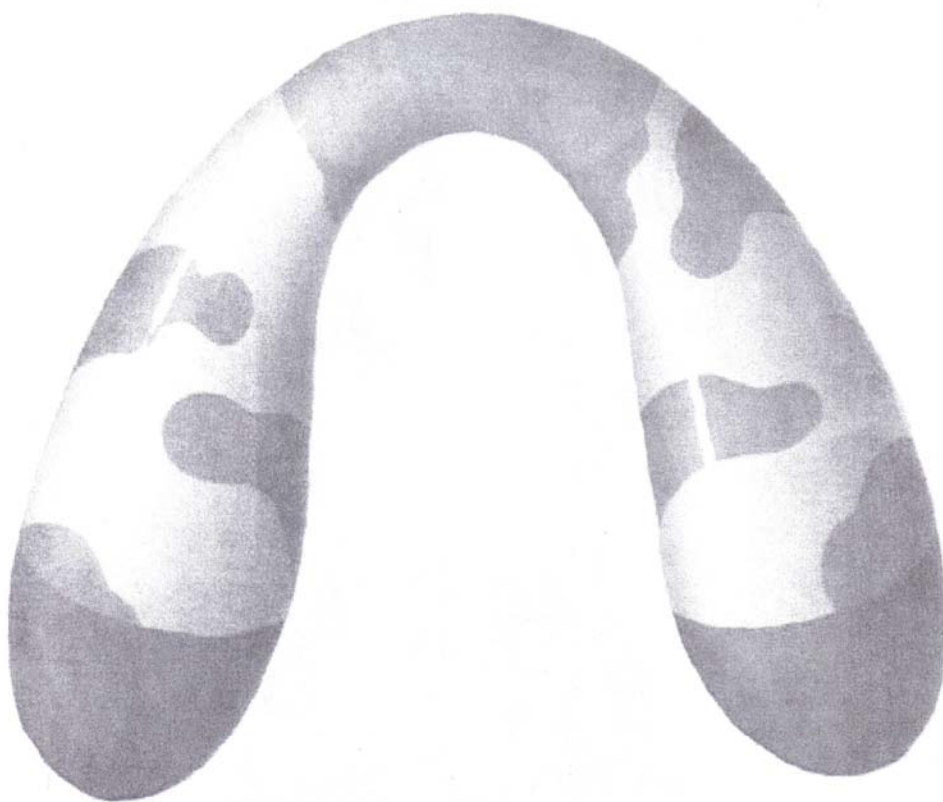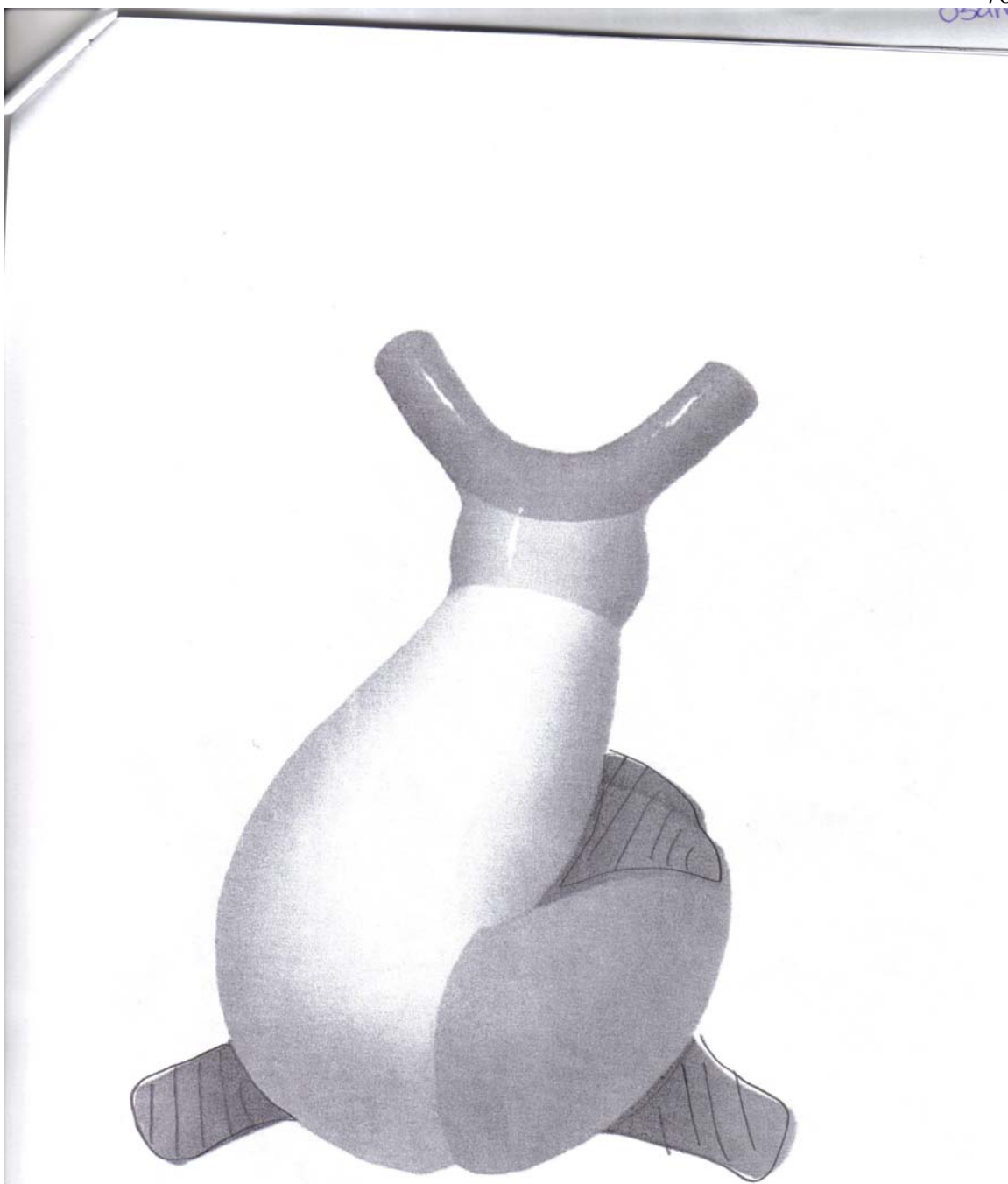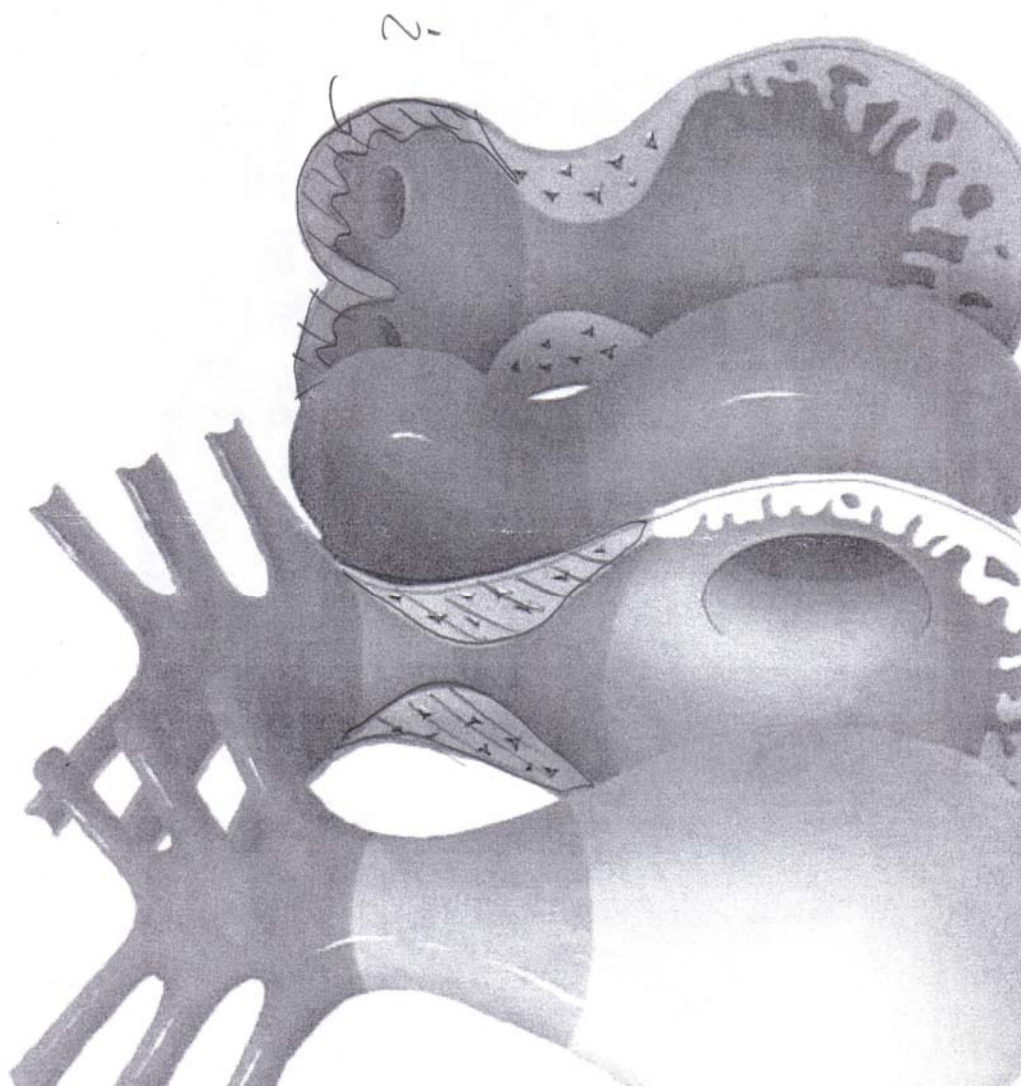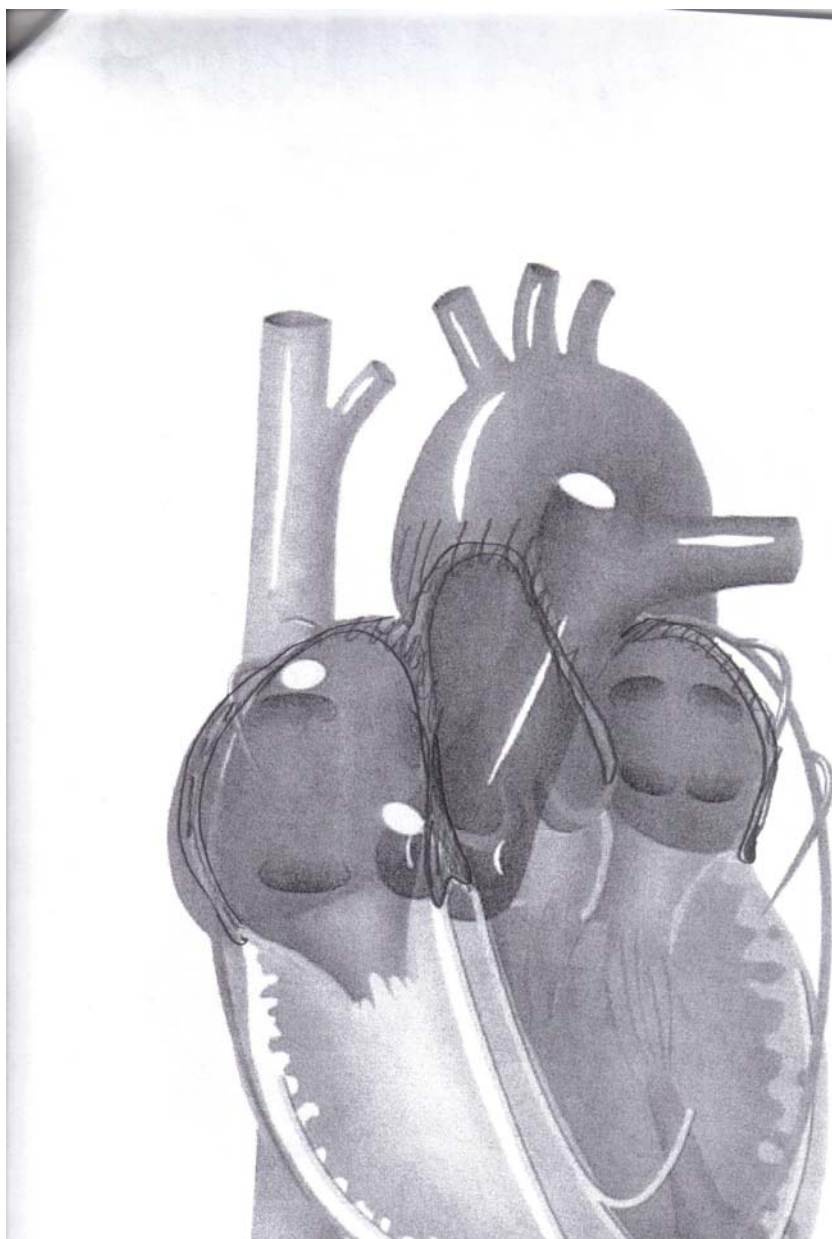
(HRT2)   pink
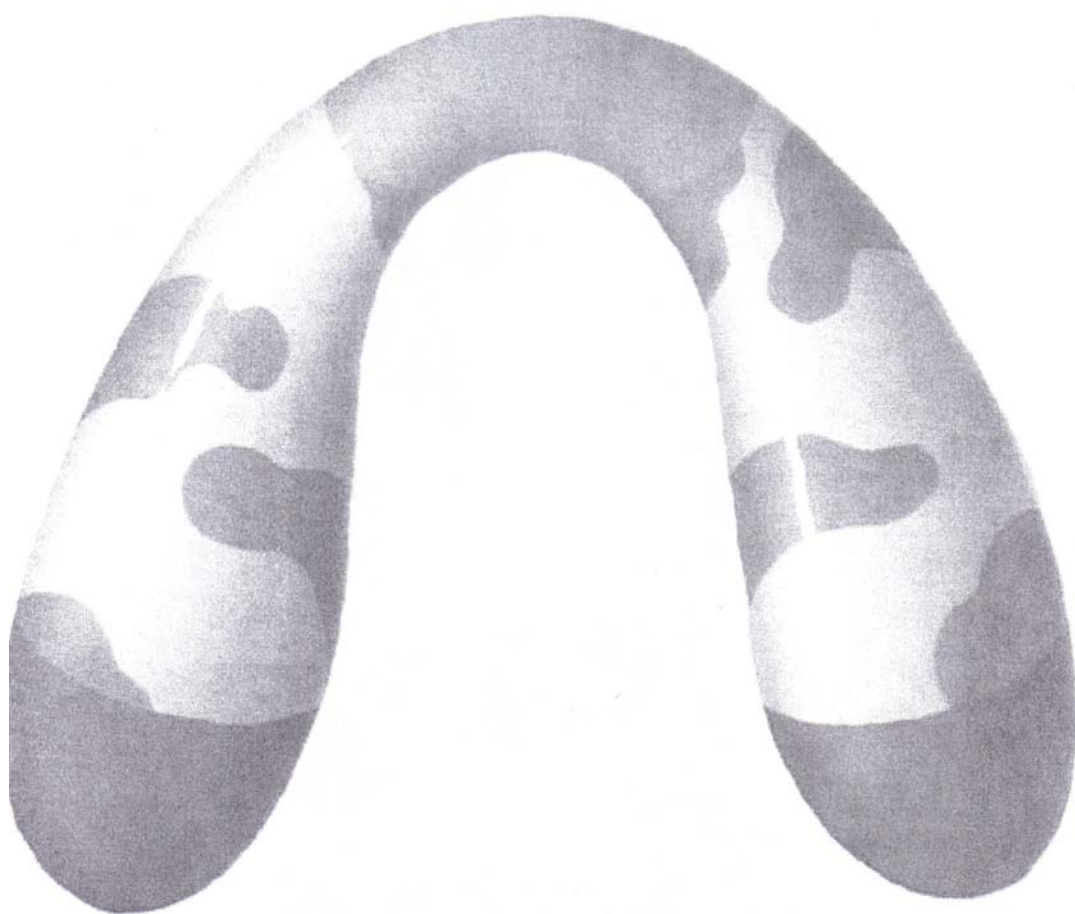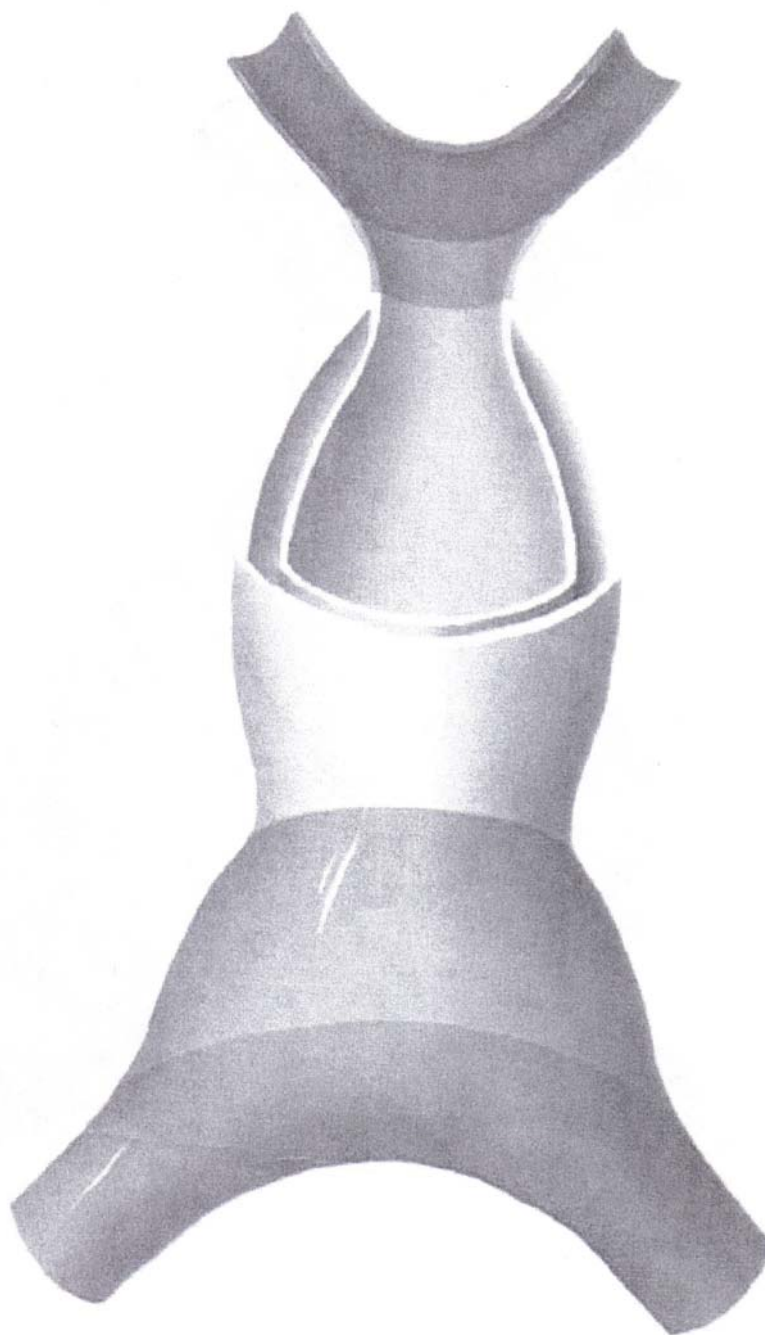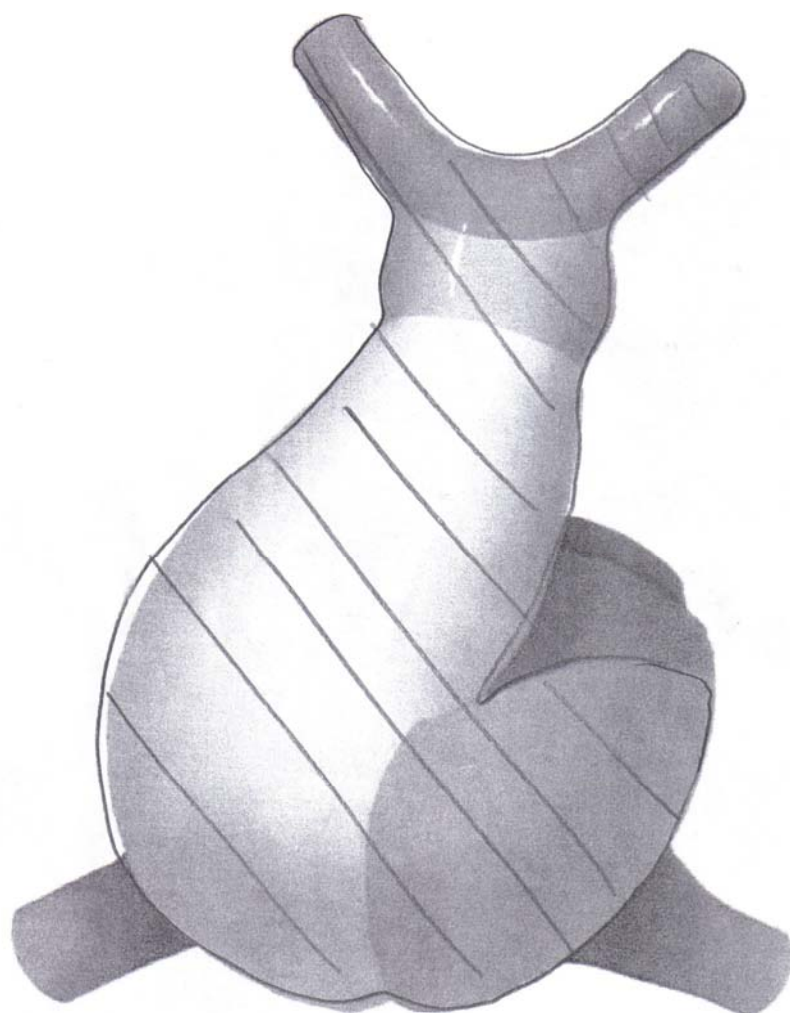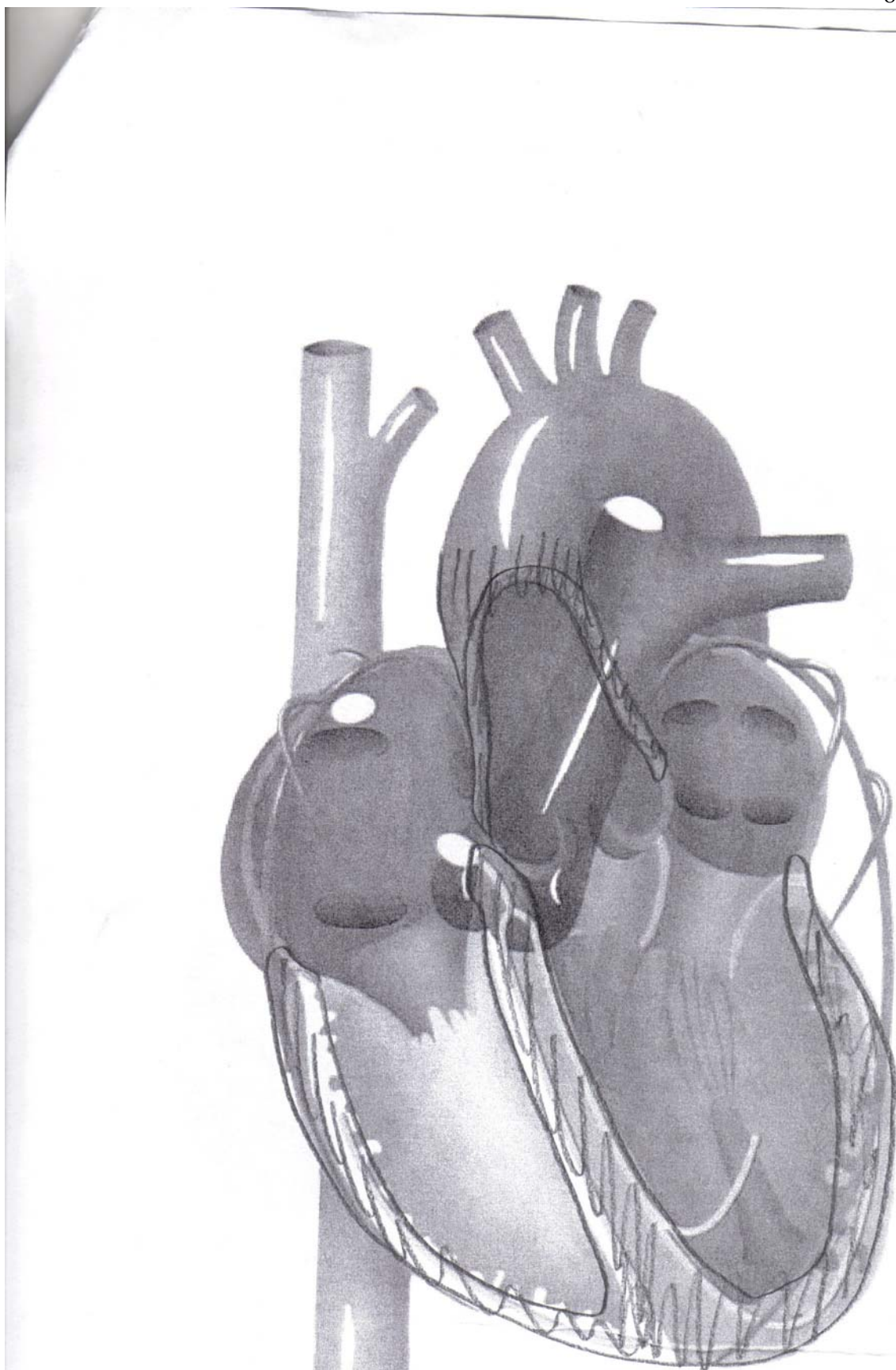
Thesis: Creating a Model 3D Animation of Heart Development

The objective of my thesis was to create a 3D animation of heart development which can be easily updated and reused when new scientific research is discovered. I created two separate videos based on the same animation model. You will see that the two animations are very similar, yet depict different aspects of heart development.

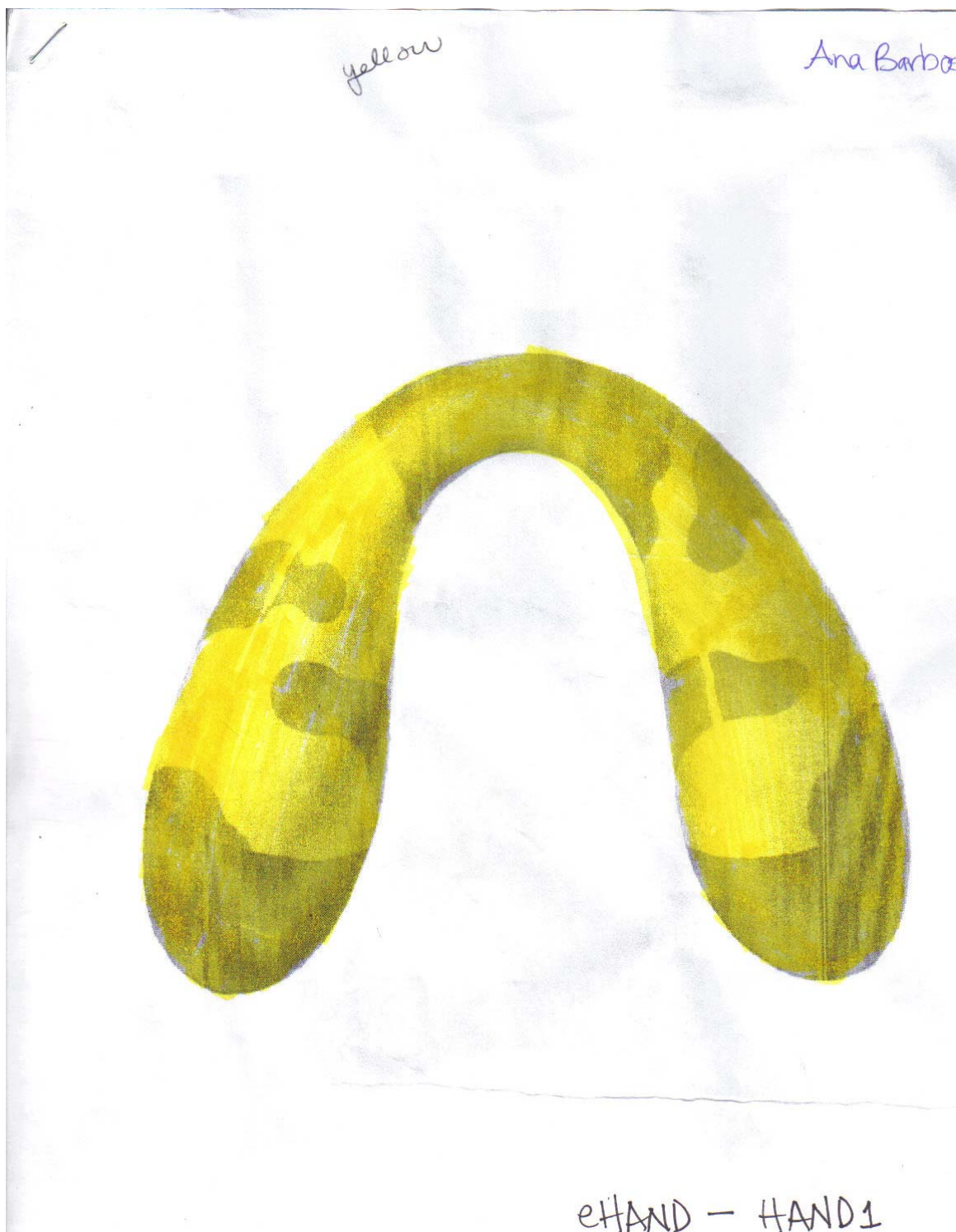I greatly appreciate your contribution to my project, and I ask that you take just a few minutes to complete the questionnaire provided.

Thank you for your time!

Ryan Carre
Department of Biomedical Communications

Cardiomyogy

Heart Development Video Questionnaire

Indicate your opinion by circling a number next to each question. **1** is least, **5** is greatest.

	Least	Most

1. How effective do you believe the first video is in depicting how the heart develops?   1 2 3 ④ 5

2. How effective do you believe the second video is in depicting the expression of transcription factors in the developing heart?   1 2 3 ④ 5

3. How helpful do you believe 3D animations are compared to 2D drawings in understanding heart development   1 2 3 4 ⑤

4. How important to you believe it is to have an animation that can be easily updated to include new research?   1 2 3 4 ⑤

5. How important to you believe it is to have an animation that can be used for multiple topics?   1 2 3 ④ 5

6. How effective was the morphing sequence in showing how the heart develops from the cardiac crescent to the adult heart?   1 2 3 ④ 5

7. How effective were the colors chosen to highlight specific areas of the developing heart?   ① 2 3 4 5
→ need to be more intense

8. How effective were the labels in the animations?   1 2 ③ 4 5

9. How appropriate was the content of the animations for an audience knowledgeable about cardiac morphology? (An answer of 5 would indicate that the animations were neither too simplistic nor too advanced for the audience.)   1 2 3 4 ⑤

10. How effective were the durations of the animations?   1 2 3 4 5
depends upon context.

What is your position at UTSW? (PI, Post-Doc, student, etc.)___PI___

Additional Comments: ___indication of Embryonic Day (Mouse + Man) would be a great improvement. A great next generation would be to be able to cross reference expression of two transcription factors at a time. a composit array of multiple factors at a given time would be more informative. This is great, I would love a copy.___

Heart Development Video Questionnaire

Indicate your opinion by circling a number next to each question. **1** is least, **5** is greatest.

		Least				Most

1. How effective do you believe the first video is in depicting how the heart develops?   1 2 3 **(4)** 5

2. How effective do you believe the second video is in depicting the expression of transcription factors in the developing heart?   1 2 3 **(4)** 5

3. How helpful do you believe 3D animations are compared to 2D drawings in understanding heart development   1 2 3 **(4)** 5

4. How important to you believe it is to have an animation that can be easily updated to include new research?   1 2 3 4 **(5)**

5. How important to you believe it is to have an animation that can be used for multiple topics?   1 2 3 **(4)** 5

6. How effective was the morphing sequence in showing how the heart develops from the cardiac crescent to the adult heart?   1 2 3 **(4)** 5

7. How effective were the colors chosen to highlight specific areas of the developing heart?   1 2 **(3)** 4 5

8. How effective were the labels in the animations?   1 2 3 **(4)** 5

9. How appropriate was the content of the animations for an audience knowledgeable about cardiac morphology? (An answer of 5 would indicate that the animations were neither too simplistic nor too advanced for the audience.)   1 2 3 4 **(5)**

10. How effective were the durations of the animations?   1 2 3 **(4)** 5

What is your position at UTSW? (PI, Post-Doc, student, etc.) _Staff_

Additional Comments: _Some of the colors were too subtle but it was very very cool._

Heart Development Video Questionnaire

Indicate your opinion by circling a number next to each question. **1** is least, **5** is greatest.

		Least				Most

1. How effective do you believe the first video is in depicting how the heart develops?   1   2   3   4   (5)

2. How effective do you believe the second video is in depicting the expression of transcription factors in the developing heart?   1   2   (3   4)   5

3. How helpful do you believe 3D animations are compared to 2D drawings in understanding heart development   1   2   3   4   (5)

4. How important to you believe it is to have an animation that can be easily updated to include new research?   1   2   3   4   (5)

5. How important to you believe it is to have an animation that can be used for multiple topics?   1   2   (3)   4   5

6. How effective was the morphing sequence in showing how the heart develops from the cardiac crescent to the adult heart?   1   2   3   4   (5)

7. How effective were the colors chosen to highlight specific areas of the developing heart?   1   2   (3)   4   5

8. How effective were the labels in the animations?   1   2   3   (4)   5

9. How appropriate was the content of the animations for an audience knowledgeable about cardiac morphology? (An answer of 5 would indicate that the animations were neither too simplistic nor too advanced for the audience.)   1   2   (3   4)   5

10. How effective were the durations of the animations?   1   2   3   (4)   5

What is your position at UTSW? (PI, Post-Doc, student, etc.) _STUDENT_____

Additional Comments: _SOME OF THE COLORS TOO SUBTLE, ADDING MORE &HUMAN_

_TIME OF DEVELOPMENT WOULD HELP._____

Heart Development Video Questionnaire

Indicate your opinion by circling a number next to each question. **1** is least, **5** is greatest.

	Least	Most

1. How effective do you believe the first video is in depicting how the heart develops? *Need to have an overall introduction on the major stages of the heart development and chronological dates*   1 2 3 ④ 5

2. How effective do you believe the second video is in depicting the expression of transcription factors in the developing heart?   1 2 3 ④ 5

3. How helpful do you believe 3D animations are compared to 2D drawings in understanding heart development   1 2 3 4 ⑤

4. How important to you believe it is to have an animation that can be easily updated to include new research?   1 2 3 4 ⑤

5. How important to you believe it is to have an animation that can be used for multiple topics?   1 2 3 4 ⑤

6. How effective was the morphing sequence in showing how the heart develops from the cardiac crescent to the adult heart?   1 2 3 4 5

7. How effective were the colors chosen to highlight specific areas of the developing heart? *Colors for certain genes are difficult to see*   1 2 ③ 4 5

8. How effective were the labels in the animations? *Need to label the main structures of heart.*   1 2 ③ 4 5

9. How appropriate was the content of the animations for an audience knowledgeable about cardiac morphology? (An answer of 5 would indicate that the animations were neither too simplistic nor too advanced for the audience.)   1 2 3 ④ 5

10. How effective were the durations of the animations? *the video 2 is little too fast to visualize the gene expression*   1 2 3 ④ 5

What is your position at UTSW? (PI, Post-Doc, student, etc.)___*PI*___

Additional Comments: ___*would be nice to acknowledge the source the research data. It is wonderful!*___

Heart Development Video Questionnaire

Indicate your opinion by circling a number next to each question. **1** is least, **5** is greatest.

	Least	Most

1. How effective do you believe the first video is in depicting how the heart develops?   1　2　3　④　5

2. How effective do you believe the second video is in depicting the expression of transcription factors in the developing heart?   1　2　3　4　⑤

3. How helpful do you believe 3D animations are compared to 2D drawings in understanding heart development   1　2　3　4　⑤

4. How important to you believe it is to have an animation that can be easily updated to include new research?   1　2　3　4　⑤

5. How important to you believe it is to have an animation that can be used for multiple topics?   1　2　③　4　5

6. How effective was the morphing sequence in showing how the heart develops from the cardiac crescent to the adult heart?   1　2　③　4　5

7. How effective were the colors chosen to highlight specific areas of the developing heart?   1　2　3　4　⑤

8. How effective were the labels in the animations?   1　2　3　④　5

9. How appropriate was the content of the animations for an audience knowledgeable about cardiac morphology? (An answer of 5 would indicate that the animations were neither too simplistic nor too advanced for the audience.)   1　2　3　4　⑤

10. How effective were the durations of the animations?   1　2　3　4　⑤

What is your position at UTSW? (PI, Post-Doc, student, etc.) _Post Doc_

Additional Comments: _very nice work that provides a very overview about the specific expression of transcription factors involved the cardiac development. This work will provide an awesome tool in order to teach both, non-cardiologists and cardiologist about cardiac development and to link basic research with clinical problems._

Heart Development Video Questionnaire

Indicate your opinion by circling a number next to each question. **1** is least, **5** is greatest.

		Least				Most

1. How effective do you believe the first video is in depicting how the heart develops? — 1 2 3 [4] 5

2. How effective do you believe the second video is in depicting the expression of transcription factors in the developing heart? — 1 2 [3] 4 5

3. How helpful do you believe 3D animations are compared to 2D drawings in understanding heart development — 1 2 3 [4] 5

4. How important to you believe it is to have an animation that can be easily updated to include new research? — 1 2 3 4 (5)

5. How important to you believe it is to have an animation that can be used for multiple topics? — 1 2 3 4 (5)

6. How effective was the morphing sequence in showing how the heart develops from the cardiac crescent to the adult heart? — 1 2 3 (4) 5

7. How effective were the colors chosen to highlight specific areas of the developing heart? — 1 (2) 3 4 5

8. How effective were the labels in the animations? — 1 2 (3) 4 5

9. How appropriate was the content of the animations for an audience knowledgeable about cardiac morphology? (An answer of 5 would indicate that the animations were neither too simplistic nor too advanced for the audience.) — 1 (2) 3 4 5

10. How effective were the durations of the animations? — 1 2 3 [4] 5

What is your position at UTSW? (PI, Post-Doc, student, etc.) _post-doc._

Additional Comments: _the second part of the movie is too simple. and it would be better if all the transcription factors are present in order, instead of showing one at one time._

Heart Development Video Questionnaire

Indicate your opinion by circling a number next to each question. **1** is least, **5** is greatest.

	Least				Most

1. How effective do you believe the first video is in depicting how the heart develops?   1 2 3 (4) 5

2. How effective do you believe the second video is in depicting the expression of transcription factors in the developing heart?   1 2 3 (4) 5

3. How helpful do you believe 3D animations are compared to 2D drawings in understanding heart development   1 2 3 4 (5)

4. How important to you believe it is to have an animation that can be easily updated to include new research?   1 2 3 4 (5)

5. How important to you believe it is to have an animation that can be used for multiple topics?   1 2 3 4 (5)

6. How effective was the morphing sequence in showing how the heart develops from the cardiac crescent to the adult heart?   1 2 3 (4) 5

7. How effective were the colors chosen to highlight specific areas of the developing heart?   1 2 (3) 4 5

8. How effective were the labels in the animations?   1 2 (3) 4 5

9. How appropriate was the content of the animations for an audience knowledgeable about cardiac morphology? (An answer of 5 would indicate that the animations were neither too simplistic nor too advanced for the audience.)   1 2 (3) 4 5

10. How effective were the durations of the animations?   1 2 (3) 4 5

What is your position at UTSW? (PI, Post-Doc, student, etc.) _faculty_____

Additional Comments: ____the transcription factor movie_____ would be more effective for a cardiology audience if all the factors were a one frame,

Heart Development Video Questionnaire

Indicate your opinion by circling a number next to each question. **1** is least, **5** is greatest.

	Least				Most

1. How effective do you believe the first video is in depicting how the heart develops? 1 2 ③ 4 5

2. How effective do you believe the second video is in depicting the expression of transcription factors in the developing heart? 1 2 3 ④ 5

3. How helpful do you believe 3D animations are compared to 2D drawings in understanding heart development 1 2 3 ④ 5

4. How important to you believe it is to have an animation that can be easily updated to include new research? 1 2 3 4 ⑤

5. How important to you believe it is to have an animation that can be used for multiple topics? 1 2 3 ④ 5

6. How effective was the morphing sequence in showing how the heart develops from the cardiac crescent to the adult heart? 1 2 3 ④ 5

7. How effective were the colors chosen to highlight specific areas of the developing heart? 1 2 3 ④ 5

8. How effective were the labels in the animations? 1 2 ③ 4 5

9. How appropriate was the content of the animations for an audience knowledgeable about cardiac morphology? (An answer of 5 would indicate that the animations were neither too simplistic nor too advanced for the audience.) 1 2 3 ④ 5

10. How effective were the durations of the animations? 1 ② 3 4 5

What is your position at UTSW? (PI, Post-Doc, student, etc.) _Research technician_

Additional Comments: _____

_____

_____

Heart Development Video Questionnaire

Indicate your opinion by circling a number next to each question. **1** is least, **5** is greatest.

		Least				Most

1. How effective do you believe the first video is in depicting how the heart develops? 1 2 3 4 **(5)**

2. How effective do you believe the second video is in depicting the expression of transcription factors in the developing heart? 1 2 **(3)** 4 5

3. How helpful do you believe 3D animations are compared to 2D drawings in understanding heart development 1 2 3 4 **(5)**

4. How important to you believe it is to have an animation that can be easily updated to include new research? 1 2 3 **(4)** 5

5. How important to you believe it is to have an animation that can be used for multiple topics? 1 2 **(3)** 4 5

6. How effective was the morphing sequence in showing how the heart develops from the cardiac crescent to the adult heart? 1 2 3 4 **(5)**

7. How effective were the colors chosen to highlight specific areas of the developing heart? 1 2 **(3)** 4 5

8. How effective were the labels in the animations? 1 2 3 **(4)** 5

9. How appropriate was the content of the animations for an audience knowledgeable about cardiac morphology? (An answer of 5 would indicate that the animations were neither too simplistic nor too advanced for the audience.) 1 2 3 **(4)** 5

10. How effective were the durations of the animations? 1 2 3 **(4)** 5

What is your position at UTSW? (PI, Post-Doc, student, etc.)___*post-doc*___

Additional Comments: ___*transcription factor animation was little too long, would be better to show expression per transcri, factor in subsequent stages.*___

The heart development movie is great! Congratulati

Heart Development Video Questionnaire

Indicate your opinion by circling a number next to each question. **1** is least, **5** is greatest.

		Least				Most

1. How effective do you believe the first video is in depicting how the heart develops? 1 2 (3) 4 5

2. How effective do you believe the second video is in depicting the expression of transcription factors in the developing heart? 1 2 3 4 (5)

3. How helpful do you believe 3D animations are compared to 2D drawings in understanding heart development 1 2 3 (4) 5

4. How important to you believe it is to have an animation that can be easily updated to include new research? 1 2 3 4 (5)

5. How important to you believe it is to have an animation that can be used for multiple topics? 1 2 3 4 (5)

6. How effective was the morphing sequence in showing how the heart develops from the cardiac crescent to the adult heart? 1 2 3 4 (5)

7. How effective were the colors chosen to highlight specific areas of the developing heart? 1 2 3 4 (5)

8. How effective were the labels in the animations? 1 2 3 4 (5)

9. How appropriate was the content of the animations for an audience knowledgeable about cardiac morphology? (An answer of 5 would indicate that the animations were neither too simplistic nor too advanced for the audience.) 1 2 3 4 (5)

10. How effective were the durations of the animations? 1 2 3 4 (5)

What is your position at UTSW? (PI, Post-Doc, student, etc.) Research Tech I

Additional Comments: _____

_____

_____

Heart Development Video Questionnaire

Indicate your opinion by circling a number next to each question. **1** is least, **5** is greatest.

		Least				Most

1. How effective do you believe the first video is in depicting how the heart develops?

   1 2 3 4 ⑤

2. How effective do you believe the second video is in depicting the expression of transcription factors in the developing heart?

   1 2 ③ 4 5

3. How helpful do you believe 3D animations are compared to 2D drawings in understanding heart development

   1 2 3 4 ⑤

4. How important to you believe it is to have an animation that can be easily updated to include new research?

   1 2 3 4 ⑤

5. How important to you believe it is to have an animation that can be used for multiple topics?

   1 2 3 ④ 5

6. How effective was the morphing sequence in showing how the heart develops from the cardiac crescent to the adult heart?

   1 2 3 4 ⑤

7. How effective were the colors chosen to highlight specific areas of the developing heart?

   1 2 ③ 4 5

8. How effective were the labels in the animations?

   1 ② 3 4 5

9. How appropriate was the content of the animations for an audience knowledgeable about cardiac morphology? (An answer of 5 would indicate that the animations were neither too simplistic nor too advanced for the audience.)

   1 2 3 4 ⑤

10. How effective were the durations of the animations?

    1 2 3 ④ 5

What is your position at UTSW? (PI, Post-Doc, student, etc.)___Research associate

Additional Comments: ___From a non scientific stand point. It would be hard to determine where the different genes are being expressed in the heart. One Suggestion. La Use the same color for the genes name as where it is expressed.

Example: HAND1 for maturation of the heart. Instead of writing HAND1 in white. Write it in yellow to indicate the expression area.

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CURRICULUM VITAE

Ryan Carre was born in Wilmington, Delaware, on September 9, 1979, the daughter of Mary Lynn Carre and Joseph J. Carre. After completing high school at Padua Academy, in Wilmington, she went on to receive her Bachelor of Science degree in biology at the College of William & Mary in Virginia, where she also minored in art and art history. During the spring semester of 2000, she studied art history at the University of St. Andrew's in Scotland. In May of 2002, she entered the Graduate School of Biomedical Sciences at the University of Texas Southwestern Medical Center at Dallas. She was awarded the degree of Master of Arts in May, 2005.

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