A Thesis for the Degree of Master of Engineering

Ultrasonic measurement of 3D geometry of the chick extraembryonic arterial bifurcation during a cardiac cycle

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GRADUATE SCHOOL

JEJU NATIONAL UNIVERSITY

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Ultrasonic measurement of 3D geometry of the chick extraembryonic arterial bifurcation during a cardiac cycle

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ABSTRACT

The features of arterial bifurcation geometry significantly affect the local changes in wall shear stress distribution that is closely related to the formation and development of atherosclerotic plaque. However, examining arterial bifurcation geometry caused by pulsatile blood flow is limited due to the difficulties in select suitable *in vivo* experimental model and methodology. The present study aims to establish a novel method on the investigation of pulsatile cyclic variation of three-dimensional arterial bifurcation geometry using the chick embryo model. A high-frequency ultrasound imaging system with a 35-MHz mechanical sector probe was used to obtain radial cross-sectional B-mode images of the chick chorioallantoic membrane arterial bifurcation. By detecting the arterial wall motion, the pulsatile cycle was observed. Then the three-dimensional arterial bifurcation geometry was reconstructed by manually segmented two-dimensional boundary data sets. Finally, the expansion ratio of the cross-section area, bifurcation angle, tortuosity, displacement of center point at the systolic and diastolic phases were measured. The result shows that the arterial wall has asymmetrical expansion and contraction during a cardiac cycle. In addition, the translational motion of the artery was founded in the chick embryo model. The high-frequency ultrasound imaging of chick extraembryonic artery model is suggested to be useful in studying of arterial pathophysiology related to arterial bifurcation geometry.

Chapter 1

INTRODUCTION

1.1 Background

As a major cause of stroke, carotid atherosclerosis usually happens at the curves and branching regions because of local generation of low wall shear stress (WSS) [1]. Therefore, the characteristics of hemodynamics in the blood vessels become important to understand the formation and development of atherosclerosis. However, the locations of atherosclerotic plaques in the region of carotid bifurcation are not accurately predicted because the local WSS highly changes with the arterial geometry parameters, such as bifurcation angle and ratio of vessel diameters [2]. Hence, the investigation of arterial structure is necessary to understand atherosclerosis development in the vasculatures.

As *in vivo* experimental animal models, rats, mice and chick embryos have been widely used for studying properties of vascular structure and angiogenesis**.** To investigate the vascular structures from animal models, many imaging technics have been widely used [2-4]. With development of the high-frequency ultrasound imaging technic, nowadays their spatial and temporal resolutions become enough to image the cardiovascular of the small animal models. Among these animal models, the chick embryo has more advantages due to the easy optical visualization and its various shapes of blood vessels [5]. Disadvantages for the chick embryo models are that the small size of arterial vessel diameter and weak vasculature.

Therefore using an ultrasound probe into the chick embryos is very difficult because of the probe size. Consider these disadvantage we used the shell-less culture chick embryo model. Due to the many environment differences compare with normal chicken embryo these models of embryo will easy to die during embryo development. Although the shell-less culture embryo has small survival rate but the chick embryo cost is lower than other animal model so it possible to use many of samples. Therefore the chick chorioallantoic membrane (CAM) model is benefit for studying geometrical motion of vascular system.

In this paper, consider the chick embryo vasculature size, we used a high-frequency ultrasound imaging system to obtain the radial cross-sectional chick CAM arterial bifurcation during a cardiac cycle. Then the vessel lumen on the CAM arterial bifurcation was segmented manually. Finally, the 3D geometry was reconstructed by these lumen boundaries and the expansion ratio of the cross-section area, bifurcation angle, tortuosity, displacement of center point at the systolic and diastolic phases were measured.

1.2 Previous studies

Numerous researchers have been attempting to understand the mechanisms of the formation and development of atherosclerotic plaques which were combined its geometry features in the carotid [6, 7] and coronary artery [8]. According to the reports, under the pulsatile flow condition, atherosclerotic plaques are usually formed in the posterior wall of the internal carotid artery. The spatial distribution of low WSS, caused by variation of blood flow, interacts with vessel geometry. Habib Samady et al. [9] reported that the low WSS segments develop greater plaques, necrotic cores and constrictive remodeling. However, on the pulsatile flow condition, the atherosclerotic plaques do not exactly coincide in the region of carotid bifurcation because the local WSS highly changes with the arterial geometry parameter differences [2].

In computational simulation of WSS of blood vessel, the wall thickness and elasticity were important parameters. However, the wall elasticity and thickness are inhomogeneous materials and the arterial wall has inhomogeneous motion by the pulsatile flow conditions. Therefore, the measurement of the wall motion during a cardiac cycle is necessary for predicting the spatial distribution of WSS. Based on this reason, Nam et al. [7] reported that the carotid arterial wall has asymmetric radial expansion and contraction in rat. However, they use a two-dimensional ultrasound imaging, but the arterial wall has complex three-dimensional motion. Therefore, Yeom et al. [2] used cross-sectional arterial imaging to reconstruct a three-dimensional geometry using automatic imaging processing and also find

the asymmetrical expansion and contraction of the wall motion.

As an animal in vivo experiment model, the chick was widely used in studying of the cardiovascular development and morphology of microvasculature due to the various vasculatures. Most previous studies on the chick circulatory system were mainly based on the static geometry or the early stage of the chick development [10] which did not consider the cyclic variation of geometry caused by pulsatile conditions. According to the advantage of various vasculatures, the chick embryo model can be used in the study of the cyclic variation of the arterial geometry.

1.3 Specific Aims

The objective of the present research is to establish an in vivo experimental model to investigate the cyclic variation of the arterial bifurcation geometry in 3D (Three-Dimension). To accomplish this objective, the aims of this research are subdivided into three as follows.

The first aim is to acquire serial cross-section B-mode images from a chick embryo model. For the reconstruct the 3D geometry, it needs that of radial cross-section image.

The second aim is to reconstruct the 3D geometry of the arterial bifurcation. For this aim, some of imaging processing technic will used to segment the B-mode ultrasound images of the arterial bifurcation.

The third aim is to analyze the wall motion of 3D geometry during a cardiac cycle. Some of geometrical parameters measured from the reconstructed 3D chick embryo arterial bifurcation geometry.

1.4 Thesis Outline

Chapter 2 provides experiment materials and imaging system. The description starts with ultrasound imaging acquisition system setup and the details about a chick embryo model. Then cross-sectional B-mode slices of the arterial bifurcation were scanned and used to successfully reconstruct 3D geometry. Finally, geometrical parameters, such as ratio of cross-section area variation, and displacement of center, bifurcation angle changing and tortuosity of the artery were computed at systolic and diastolic phases.

Chapter 3 results of 3D geometry at systolic and diastolic during a cardiac cycle. The 3D arterial wall motion was observed. Compare two different embryo sample, observed different wall expansion and contraction motion. In addition, the displacement centerline was found.

Chapter 4 discusses cyclic variation of the 3D geometry. The wall motion in the CAM artery represents two kind of the wall motion. One is the translational motion and the other motion is expansion and contraction.

Chapter 5 concludes present study, and suggests future studies.

Chapter 2

MATERIALS AND METHODS

2.1 Shell-less embryonic culture

The chicken eggs were incubated in a digital incubator (RCOM PRO 50, Autoelex Co., Korea) at 37℃ and 60% humidity. After 3 days of incubation, the contents of embryonic eggs were removed from its shell and placed on a petridish (Diameter of 90 mm, Thickness of 20 mm). The shell-less embryonic culture consists of the following steps (shown in the Figure 2.1):

i. Erect eggs on the egg case around 10 minutes after 3days of incubation.

ii. Remove its shell carefully and place on a petridish.

iii. Place the petridish with the egg contents on the incubator..

Until at the day of 11 of incubation, the chick extraembryonic chorioallantoic arteries will develop as a reasonable size of an experiment model for study arterial wall movement using an ultrasound image system. In the development of shell-less chick embryo culture, the embryos will easy to die due the many environment differences compare with normal chicken embryo. Therefore in our experiment, at the 11 days of the embryo development, around 4 embryos will successfully live from 40 embryo samples. All experimental procedures were approved by the Ethics Committee of Jeju Ntational University.

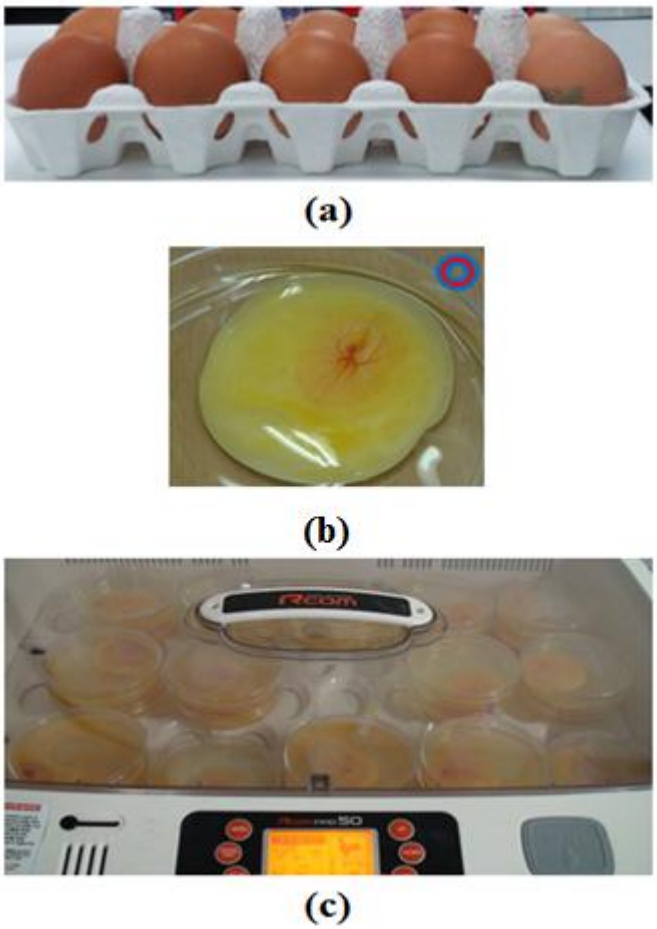

Figure 2.1. Procedures of shell-less culture. (a) Erect eggs on the egg case around 10 minutes. The air cell which was an empty space located at the large end of the egg, should be placed in upward direction. In this way the chorioallantoic membrane will move to the up direction then it can be easy to remove its shell and didn't hurt the chorioallantoic vasculatures. (b) Shows the shell removed chick embryo sample in a petridish at 3 day of the development. (c) Place the shell-less embryo contents in petridishes on the incubator (RCOM PRO 50, Autoelex Co., Korea).

2.2 High-frequency ultrasound imaging system

A personal computer based high-frequency ultrasound imaging system was used to this experiment. The imaging system was configured by a probe (SN21141, Capistrano Labs Inc., San Clemente, CA, USA) including a 35 MHz broadband acoustic transducer (35_{Ti}MHz, Capistrano Labs Inc., San Clemente, CA, USA) and an ultrasound imaging board (PCB v4.2, Capistrano Labs Inc., San Clemente, CA, USA). The probe was also equipped with a nosepiece (PN18923, Capistrano Labs Inc., San Clemente, CA, USA) and a cover film (RB820, Capistrano Labs Inc., San Clemente, CA, USA). Figure 2.2 shows the schematic of a computer based high-frequency ultrasound imaging system and the conceptual diagram of the experiment set up. The parameters of imaging system are shown in the Table 2.2. Image acquisition of the system was set at a frame rate of 30 fps. The scanning angle of probe was set to ±10°. The distance from the transducer pivot to the transducer surface was 10 mm and the distance from the transducer surface to the probe surface was 1 mm. The central frequency was 35 MHz, the max of amplitude was 127.5 and the loop gain was 60 dB.

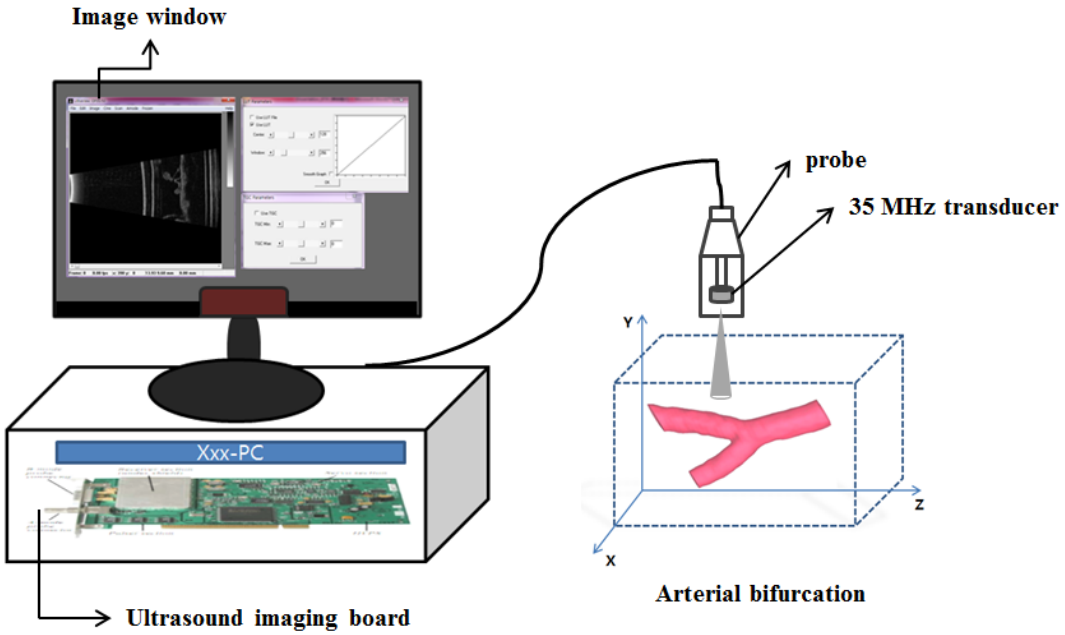

Figure 2.2. The computer based ultrasound imaging system and the conceptual diagram of the experiment set up. A mechanical sector probe (SN21141, Capistrano Labs Inc., San Clemente, CA, USA) with a 35 MHz transducer (35_{Ti}MHz, Capistrano Labs Inc., San Clemente, CA, USA) was connected an ultrasound imaging board (PCB v4.2, Capistrano Labs Inc., San Clemente, CA, USA). The probe placed on the subject like a figure, the arterial bifurcation will show in the image window. Through the XYZ positional movement, the radial cross-section artery was scanned by the sector probe.

Table 2.1. Parameters of the high-frequency ultrasound imaging system

Parameter	**Value**
Frame rate	30 fps
Scanning angle	±10º
Transducer pivot to transducer surface	10 mm
Transducer surface to probe surface	1 mm
Central frequency	35 MHz
Amplitude	127.5
Loop gain	60 dB

2.3 In vivo measurement

In order to apply the imaging probe to the subject, we used an acoustic medium layer. A mineral oil (Alfa Aesar, Ward Hill, USA) was used to put on the egg contents. The chick CAM is very weak so the probe cannot directly contact it. Therefore the oil can be a medium layer and also protect the membrane from braking by attrition with the probe. Then a subject was placed on an animal imaging stage with an XYZ positioner. Before process the in vivo experiment, the room temperature was set at around 28℃ by air conditioner. Focal zone of the probe was fit to the appropriate position of the radial CAM artery. While the transducer was on sectorial motion with a scanning angle range, 256 A-mode scan lines were obtained in one frame for a B-mode image. The image was acquired at a frame rate of 30 fps. One slice was composed of 90 frames so the time of data at one slice was 3 s. To obtain the multi-slice images of the CAM arterial bifurcation, the probe was scanned along the longitudinal direction of the artery at the interval of 0.1 mm between two consecutive slices. The scan direction is shown in Figure 2.3. The figure is a photo image of the 11 days of chick embryo development. The scan direction, CAM artery and vein are marked. Total of 40 slices of the cross-sectional arterial images were acquired along the scan direction in each embryo samples (data of 2 representative samples were obtained in this experiment).

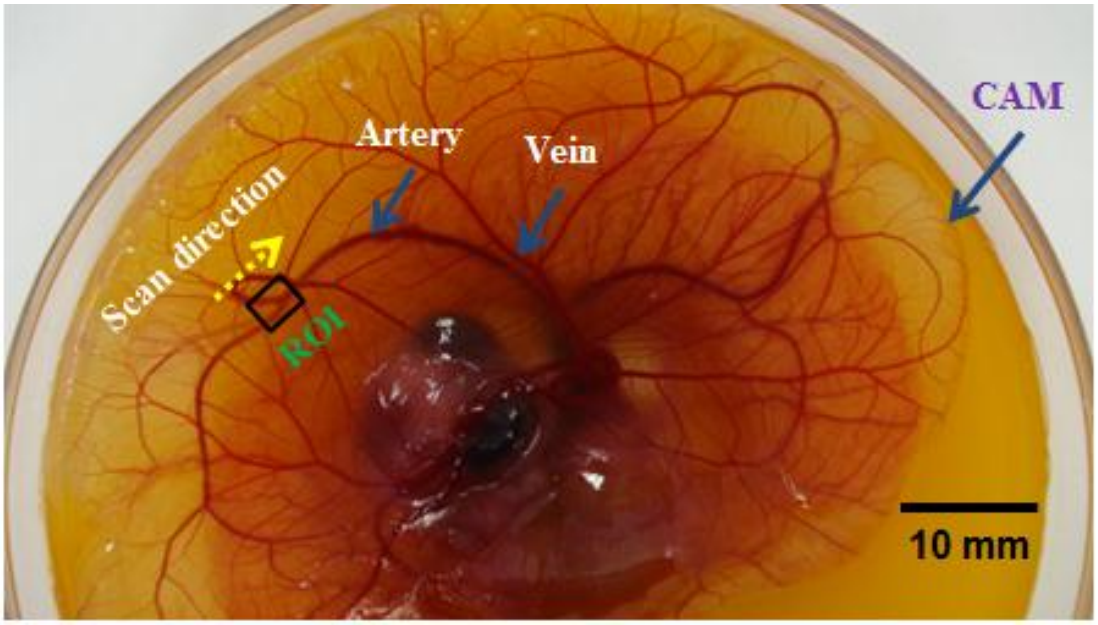

Figure 2.3. The photo image presents an 11 day chick embryo development. The region of interest (ROI) with the scan direction is marked in the left side of figure. The CAM artery and vein are marked with blue color arrows. The color of artery looks darker than that of the vein.

2.4 Data analysis

2.4.1. 3D lumen geometry reconstruction

The 3D geometry reconstruction method is based on a two-step approach:

A. Systolic and diastolic phases were detected.

B. The lumen boundaries were manually detected.

A. Systolic and diastolic phase detection

Figure 2.4 (a) shows the radial cross-sectional images of the CAM arterial bifurcation. The artery and surrounding tissues are marked with the blue arrows. The cyclic variations of the arterial wall displacement could be obtained from the time domain plotting of the arterial wall motion using Matlab (MathWorks, Natick, MA, USA). The line is the arterial cross-section marked with the yellow dotted line in the Figure 2.4 (a). The result is presented in Figure 2.4 (b). Systolic and diastolic phases are detected and marked in the picture. The red dot line represents the systolic phase and the blue dot line represents the diastolic phase. In cardiovascular system, when hearts contract, the blood vessel will expand due to the accelerated blood flow. Oppositely when the hearts expand, the blood vessel will contract. Therefore, the blood vessel will expand quickly and contract slowly during a period of heart beat. This phenomenon was found from the chicken embryo. Through the time domain plotting the arterial wall displacement can be shown in Figure 2.4 (b). The systolic to diastolic phases well recognized based on this phenomenon.

B. The lumen boundary detection

The manual segmentation in this study was performed by using an imaging processing software Amira (FEI visualization sciences group, Dahlem, Berlin, Germany). First, select several points around lumen boundary by the painting tool in the software. Then the points were connected using a small circular brush and the entire lumen area was painted based on the connected circles. Finally, these images were taken 3D volume rendering and the boundary edge was extracted. Figure 2.5 (a) shows the segmentation process and (b) shows results of segmentation at three different slices. Using these synchronized segmentation data, finally the 3D geometry was successfully reconstructed by “surfacegen” function in Amira software.

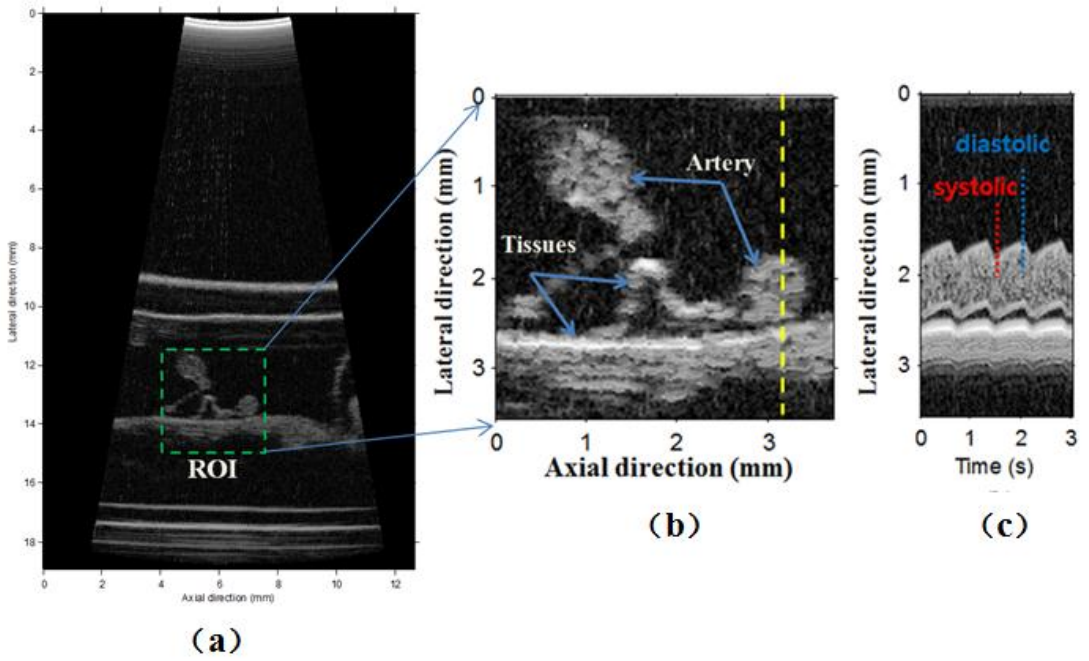

Figure 2.4. (a) Shows the frame image of the first slice at the diastolic. Artery and tissues are brighter than the surrounding environment. Which are shown in (b). In cardiovascular system, if hearts contract, the blood vessel will expand due to the accelerated blood flow. Oppositely when the hearts expand, the blood vessel will contract. Therefore, the blood vessel will expand quickly and contract slowly during the period time heartbeat. Arterial wall displacement is obtained from the time domain plotting of the arterial wall motion. As shown in (c).

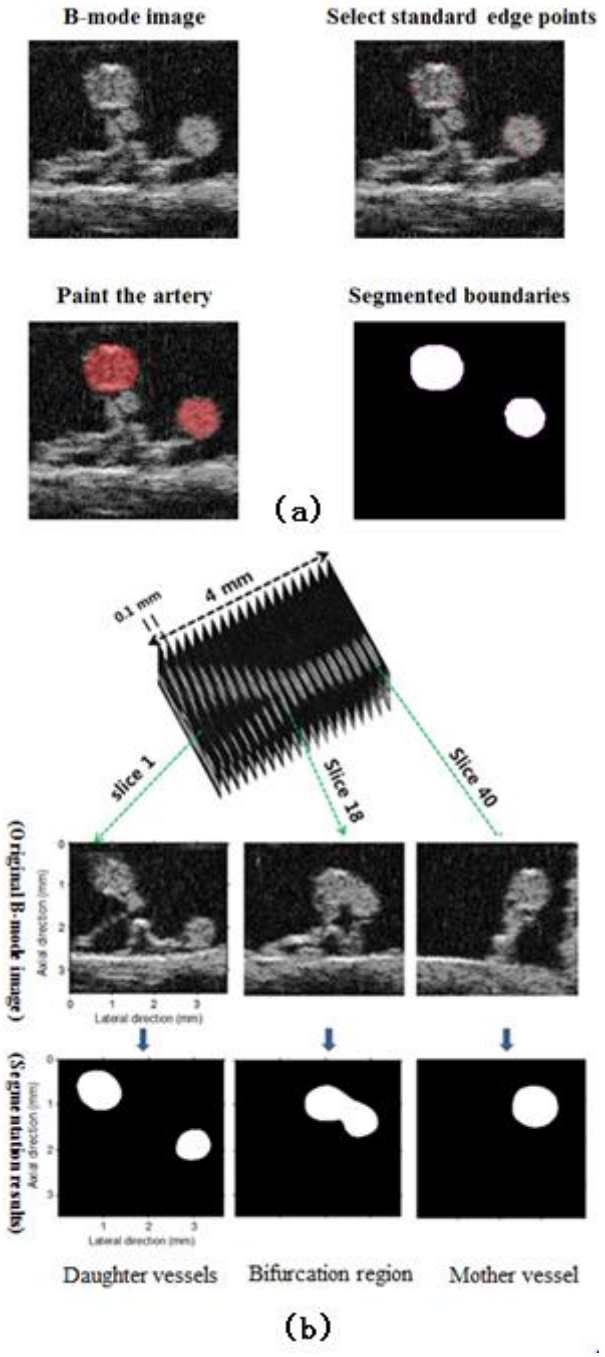

Figure 2.5. (a) Shows the manual segmentation imaging processing procedures. The three typical different case of the segmentation results at slice1, slice18 and slice 40 which were called daughter vessel, bifurcation region and mother vessel are shown in (b).

2.4.2. 3D lumen center point extraction from the lumen boundary data set

Figure 2.6 shows a procedure of lumen center points tracking method. The procedure consists of following three parts.

The first part is case classification. The boundary has one closed boundary and two closed boundary. The center of mass was defined as a center point. For the two closed boundaries, two center points were extracted using the “regionprops” function in Matlab software (MathWorks, Natick, MA, USA).

The second part is ellipse fitting which was based on the ratio of major axis of minor axis. For the one closed boundary, if the ratio is $R<1.4$, that will extract one center of mass. The ratio R value was manually determined according to the consecutive radial cross-section lumen shape. If the ratio is $R>1.4$, the closed boundary need to segment two parts of closed boundary by a cutting line.

The third part is finding the cutting line. The boundary which was $R>1.4$, will divide to two parts by the major axis. Then the two points which make the shortest displacement which was from the center of major axis to boundary edge were connected and make a straight line. That line was called be a cutting line. Using this cutting line, one closed boundary was segmented two closed boundary and the two center points were extracted.

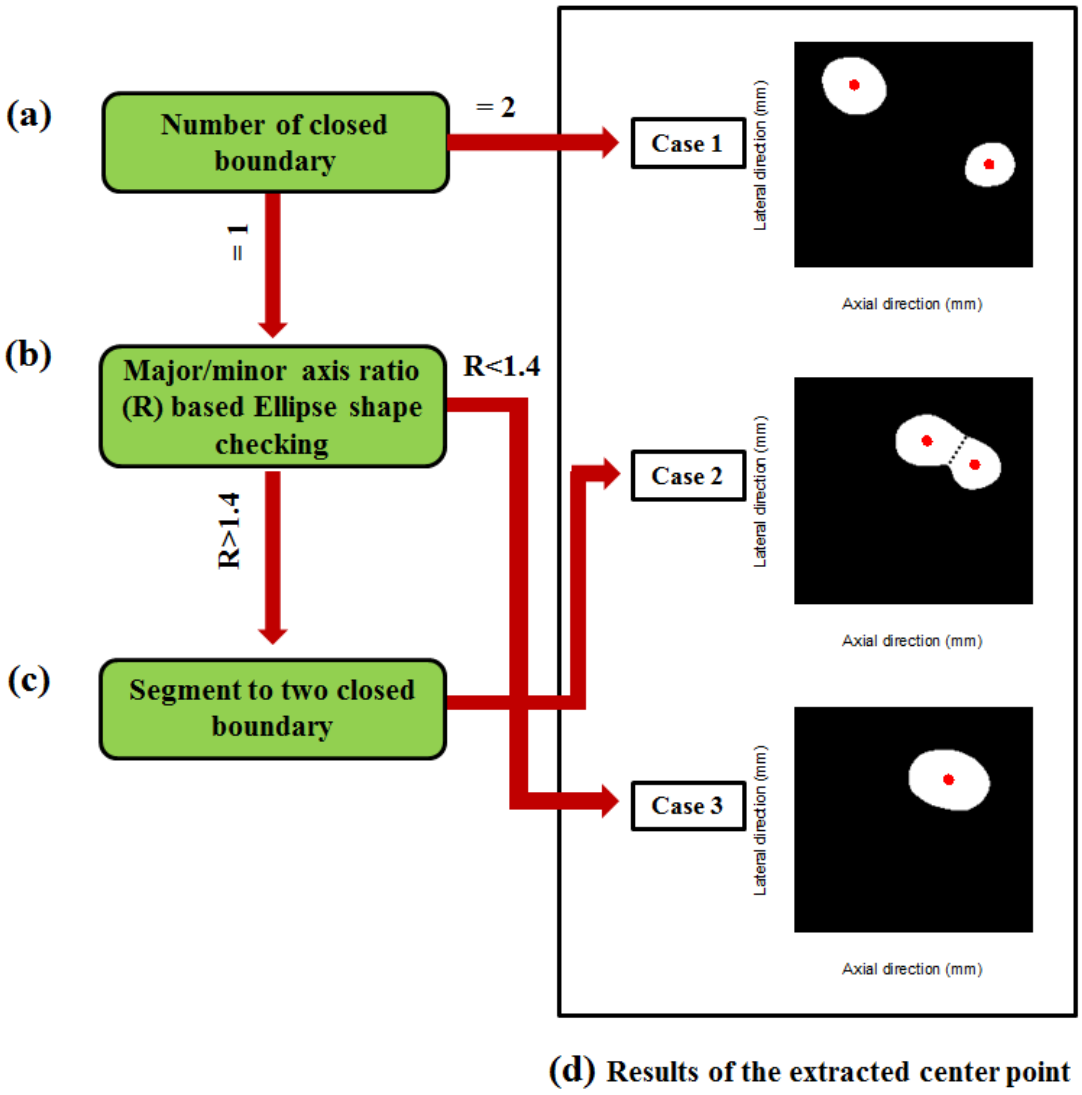

Figure 2.6. Schematics of the center point extraction. (a) shows center points extraction method which was for two closed boundaries. (b) The ratio of major axis of minor axis value based ellipse fitting. (c) One closed boundary which was R>1.4 segmented to two region boundaries by cutting line. (d) The results of the extracted center point from three different case of boundaries are marked with red color dot.

2.4.3. Quantitative analysis

A. Expansion ratio of the cross-sectional area

For comparison of lumen area at the systolic and diastolic phases, the cross-sectional areas were averaged along the longitudinal direction. The upstream artery is marked as section A and the two artery branches in the downstream are marked as section B and section C in left side and right sides, respectively. Figure 2.7 shows reconstructed 3D CAM arterial geometry at a systolic phase. The each divided section is marked in the figure. In each section, 14 slices of the cross-section areas were averaged at systolic and diastolic phases.

B. Displacement of center points

The displacement is the distance between the center points at systolic and the center points at diastolic phases. The displacement of the center points also divided three sections and 14 slices of the displacement were averaged in each section.

C. Bifurcation angle

The Figure 2.7 (b) shows schematic of bifurcation angle calculation method. The angle was calculated by scalar product formula:

$$b \cdot c = ||b||\,||c||\,\cos\theta$$

The vector $\vec{b}$ is an averaged value which was calculated from bifurcation region point O to the end point B. The vector $\vec{c}$ is an averaged value which was calculated from bifurcation region point O to the end point C. Then took these two vectors $\vec{b}$ and $\vec{c}$ to brought into the scalar product formula.

D. Tortuosity

The tortuosity was calculated by following formula [11]:

$$\text{Tortuosity} = \frac{\mathrm{L}-\mathrm{d}}{\mathrm{d}} = \frac{\sum_{\mathrm{j}=1}^{\mathrm{n}} \sqrt{\left(\mathrm{x_j}-\mathrm{x_{j-1}}\right)^2+\left(\mathrm{y_j}-\mathrm{y_{j-1}}\right)^2+\left(\mathrm{z_j}-\mathrm{z_{j-1}}\right)^2}}{\sqrt{(\mathrm{x_n}-\mathrm{x_0})^2+(\mathrm{y_n}-\mathrm{y_0})^2+(\mathrm{z_n}-\mathrm{z_0})^2}} - 1$$

One L is a length of curve which was from starting point A to the end point B and it called A-B tortuosity. The other one is a length of curve which was from starting point A to the end point C and it called A-C tortuosity. x_j, y_j and z_j are the position value of the center point. Parameter d is a length of line which was from starting point A to the end point B or C. Took these parameters to this formula the tortuosity of A-B curve and A-C can be calculated.

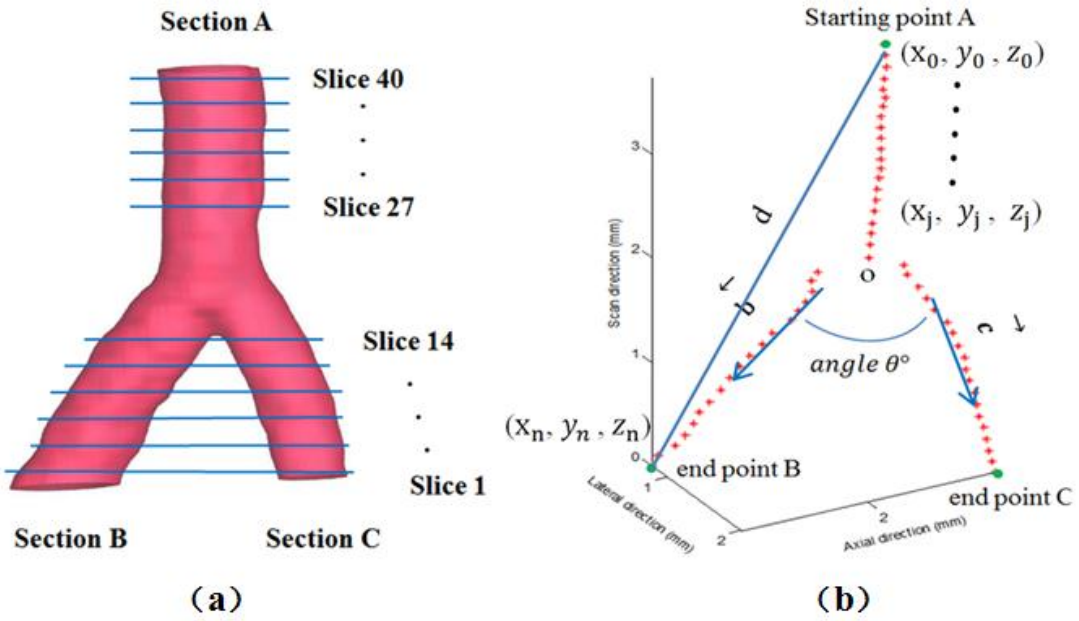

Figure 2.7. (a) Shows a 3D reconstructed CAM arterial bifurcation at a systolic phase. The lumen area of 14 slices in the each section was averaged. The bifurcation angle was calculated using the scalar product formula. The vectors of *b* and *c* are the averaged vectors in section B and section C respectively. Tortuosity is to relate the length of the vessel segement to the straight-line (AC) distance between its starting point to its end point. These parameter are shown in (b).

Chapter 3

RESULTS

3.1 Systolic and diastolic geometry

Figure 3.1 shows a representative result of the reconstructed 3D CAM arterial bifurcation geometry. The systolic and diastolic phases are superimposed on a single frame, in order to see the wall motion between the two phases. Longitudinal variation of the wall displacement and the radial wall motion downstream and upstream are well recognized. In the sample #1, the artery shows translational vessel movement to up- and down- direction. In figure (b), the section B moves right but section C moves up and down direction. It means the translational motion is different at each section. The sample #2 shows another 3D CAM arterial bifurcation geometry obtained from a different *shells-less embryonic* samples at systole and diastole phases. In this sample, the arterial wall motion in the downstream, section B and C moved to opposite direction, one right and the other left. The results show regional variation in wall movement direction depending on branch section and its curvature geometry.

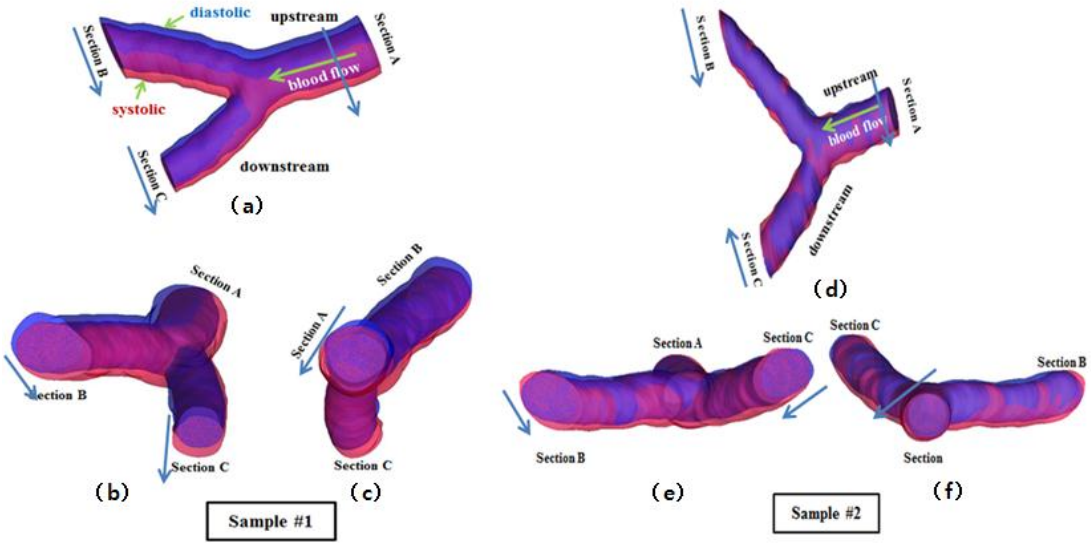

Figure 3.1. Representation of the reconstructed 3D CAM arterial bifurcation geometry for sample #1 (a-c) and sample #2 (d-f). (a) & (d) are longitudinal–section and (b) & (e) are cross-section, and (c) & (f) are of radial artery at downstream.

3.2 Variation of the centerline during pulsatile cycle

The centerline of the artery is represented with the 3D reconstructed bifurcation geometry in Figure 3.2. The red and blue dots represent systolic and diastolic center points respectively. The center line variation in sample #1 shows that the artery mainly moves right direction but in sample #2 the artery moves depending on the section. In the sample #2, centerline of section A and B move to right direction but that of section C moves to left direction. When compared with sample #1, the arterial displacement shows less changing in center movement in the sample #2. This phenomenon may be caused by the geometrical effects, such as bifurcation angle or branch curvature, and also related with the local elasticity of the vessel wall requiring clear understanding in the future studies.

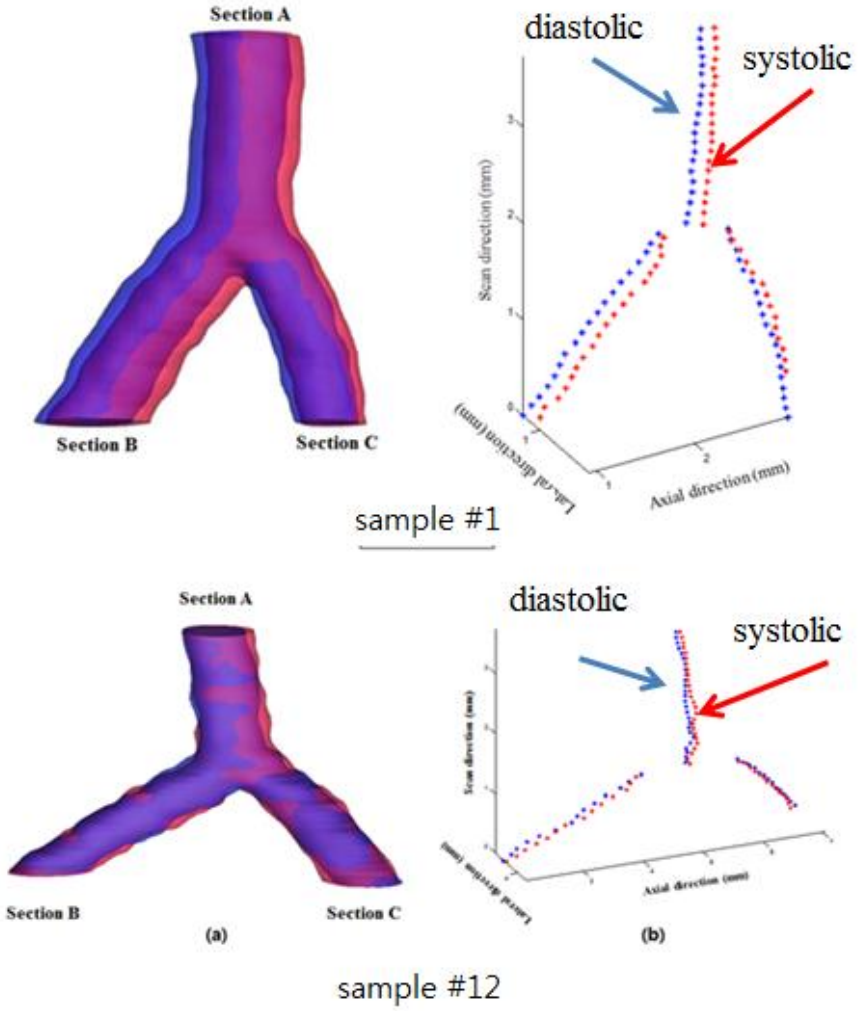

Figure 3.2. The extracted centerlines are presented. Considered the terms of mother branch, and two daughter branches. The red dots show a systolic phase and the blue dots show a diastolic phase.

3.3 Characteristics of cyclic variation of the bifurcation geometry

The geometrical parameters, such as ratio of cross-section area, displacement of center, bifurcation angle and tortuosity of the artery, were computed. The results are tablet shown in Table 3.1.

In the case #1, the expansion ratio was 18.8 for section B, which was larger than those in sections A and C. The displacement of center point in section B was 0.162 mm that was also larger than those in other sections. However, the large expansion ratio was not always with largest displacement. In the case #2, the largest expansion ratio was shown in the section C but the displacement in section C was not largest one. The three sections of two cases showed area expansion. In these two cases, the expansion ratio was unexpectedly large up to 10 to 20 except the section C of the case 2.

The bifurcation angle in the case 1, was 51.95° at a diastolic phase and 52.09° at a systolic phase. In this case the bifurcation angle was not much changed during a pulsatile cycle. In case 2, bifurcation angle were 100.41° at diastole and 96.32° at systole. In this case the bifurcation angle was changed more than 4°. These angle variations of bifurcation indicate that two sections of the branch moved to opposite direction. Figure 3.1 (b) shows the direction of the branch movement between diastole to systole.

In addition, tortuosity of the branch curves was calculated. The tortuosity value in the large bifurcation angle was large but not much changing during diastolic and systolic phases.

Table 3.1. The variation of bifurcation parameters at systolic and diastolic.

Parameters	**Sample (#)**	**Phase**	Section A	Section B	Section C
Expansion ratio of cross-sectional area (%)	Case 1	Systolic to diastolic	14.8	18.8	14.6
	Case 2	systolic to diastolic	17.2	11.8	27.9
Displacement of center point (mm)	Case 1	Systolic to diastolic	0.126	0.162	0.108
	Case 2	systolic to diastolic	0.054	0.108	0.090
Bifurcation angle (°)	Case 1	Diastolic	51.95°		
		Systolic	52.09°		
	Case 2	Diastolic	100.41°		
		Systolic	96.32°		

			A-B	A-C
Tortuosity	Case 1	Diastolic	0.0715	0.1001
		Systolic	0.0952	0.0759
	Case 2	Diastolic	0.2745	0.1579
		Systolic	0.2773	0.1644

Chapter 4

DISCUSSION

The main goal of the present study is to establish an *in vivo* experiment model to investigate the cyclic variation of the arterial bifurcation geometry in 3D. For this purpose, the chick embryo was chosen as an appropriate model. The advantage of the CAM artery is the various geometry suitable to study geometrical changing of arterial bifurcation caused by pulsatile blood flow. The geometry variation of the artery was obtained by a high-frequency ultrasound imaging system and through imaging processing techniques the 3D arterial geometry was successfully reconstructed. The results show that the ultrasonic measurement in the spatio-temporal variations of arterial geometry during a cardiac cycle could be obtained from CAM artery at an 11 -day of chick embryo development.

Some of the geometrical parameters such as bifurcation angle, centerline displacement of arteries, and tortuosity were calculated. The results indicate that the arterial translation direction could be obtained from these parameters during a cardiac cycle. Tortuosity may be a possible risk factor for an arterial disease [11] that the value of our measurement may help the study in cardiac vascular diseases using a chick embryo model.

According to recent studies in our laboratory, the rat carotid artery bifurcation expands and contracts asymmetry during a cardiac cycle [7]. The same phenomenon was found in the chick embryo CAM arterial bifurcation model. The pulsatile variation of the geometry may

affect the local WSS change that is one of the important factors for cardiovascular diseases.

In this study the translation of the wall motion was founded to be large. The CAM artery was placed on the liquid without surrounding tissues. When the blood is flowing and hemodynamic force is acting on the geometry, causing the arterial wall translate. If the artery is in the surrounding bounded tissues, like the artery of the rat, the tissue will impede the arterial translation. Then the energy from the blood flow transferred to the tissue. In the CAM model, both the transitional and expanding wall motions were measured. These combined wall motions will be seen in the coronary artery movement caused by the pulsatile blood flow and heart motion. Therefore this chick CAM may provide an animal model for studies on the coronary vascular diseases such as coronary atherosclerosis.

The present study has a number of limitations. The shell-less *in vivo* model was different from the normal embryo. There are many different factors, such as air, room temperature and easy bacterium infection. Some of CAM arteries were placed in the deeper position but the short focal length with shallow DOF of the HFUS transducer couldn’t reach to those vessels. The resolution of image was also limited. Another limitation in this experiment was that the CAM artery was very weak so it couldn’t directly contact the probe, requiring an acoustic medium. The mineral oil was put on the CAM so that the liquid will give a pressure to the artery which may affect the pulsatile condition. Further advances in ultrasound image analysis technique are required to establish a suitable automatic segmentation technique. For the statistically analysis, it needs to take more cases of chick

embryo for investigating the pulsatile variation of arterial bifurcation geometry.

Chapter 5

CONCLUSION AND FUTURE STUDIES

5.1 Conclusion

In the present research, we successfully achieved observation of the variation of 3D geometry during a cardiac cycle in a chick embryo model. The results show that the artery has asymmetrical expansion and contraction during a cardiac cycle. In addition, the translation motion of the artery was found to be large. These findings will be useful information to the estimation of local changes in WSS distribution in the arterial bifurcation. And this chick embryo model may provide a new animal model to study arterial pathophysiology.

5.2 Future studies

Currently, the 3-D geometry reconstruction was used by manual segmentation so it needs an automatic technique to extract lumen from ultrasound images. The high temporal observation of the arterial motions during a whole cardiac cycle would be obtainable.

For the statistical analysis, more cases of chick embryo were needed for the investigating of the cyclic variation of the arterial bifurcation geometry caused by pulsatile blood flow.

According to the advantage of easy visualization, the arteria bifurcation motion can be

investigated in the chick embryo using a microscopic. Then the results of ultrasound imaging and microscopic image will be compared quantitatively for more accurate analysis of the wall motion.

Finally, the quantitative observation of the various 3-D geometries during a cardiac cycle can be used as input parameters and confirmation tools in computer simulation of the hemodynamic characteristics for prediction of the atherosclerosis plaque.

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