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Bulk Flow and Near Wall Hemodynamics of the Rabbit Aortic Arch and Descending Thoracic Aorta: A 4D PC-MRI Derived Computational Fluid Dynamics Study

Animal models offer a flexible experimental environment for studying atherosclerosis. The mouse is the most commonly used animal, however, the underlying hemodynamics in larger animals such as the rabbit are far closer to that of humans. The aortic arch is a vessel with complex helical flow and highly heterogeneous shear stress patterns which may influence where atherosclerotic lesions form. A better understanding of intraspecies flow variation and the impact of geometry on flow may improve our understanding of where disease forms. In this work, we use magnetic resonance angiography (MRA) and 4D phase contrast magnetic resonance imaging (PC-MRI) to image and measure blood velocity in the rabbit aortic arch. Measured flow rates from the PC-MRI were used as boundary conditions in computational fluid dynamics (CFD) models of the arches. Helical flow, cross flow index (CFI), and time-averaged wall shear stress (TAWSS) were determined from the simulated flow field. Both traditional geometric metrics and shape modes derived from statistical shape analysis were analyzed with respect to flow helicity. High CFI and low TAWSS were found to colocalize in the ascending aorta and to a lesser extent on the inner curvature of the aortic arch. The Reynolds number was linearly associated with an increase in helical flow intensity ($R = 0.85$, $p < 0.05$). Both traditional and statistical shape analyses correlated with increased helical flow symmetry. However, a stronger correlation was obtained from the statistical shape analysis demonstrating its potential for discerning the role of shape in hemodynamic studies.

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Introduction

Animal models have been widely used to gain a better understanding of the mechanisms involved in atherosclerotic plaque formation. These models offer an opportunity to investigate mechanistic pathways through gene or surgical manipulation. Small animals such as mice and rats have been most commonly used due to the ease of surgical or genetic alterations and short time frame for disease formation [1]. However, their small size results in a hemodynamic environment far removed from that of humans. For instance, wall shear stresses (WSS) in mice have been reported to be approximately 20 times greater than in humans [2]. Larger animals such as pigs and rabbits are employed less commonly due to their significant expense, though the vessel caliber and resulting hemodynamics are much closer to that of humans. In particular, the rabbit aorta has been used to investigate the association between WSS and atherosclerotic lesion formation [3,4]. These previous studies primarily focused on the descending aorta and did not study the intraspecies variation in aortic arch flow.

Image-based flow modeling using computational fluid dynamics (CFD) requires accurate geometry and either measurements or valid assumptions on the flow entering and leaving the vessels of interest in order to provide accurate boundary conditions. Prior studies of rabbit hemodynamics have made various assumptions including steady flow [3,4], sinusoidal waveforms [5], and neglecting out-of-plane ascending aorta inlet velocities [6]. Highlighting the importance of including the full three-dimensional (3D) velocity profile at the aortic inlet, Morbiducci et al. showed that neglecting this can result in a misleading representation of human aortic hemodynamics [7]. Four-dimensional phase contrast (PC) magnetic resonance imaging (MRI) provides the ability to measure all three velocity components and their temporal variation. Coupling these measurements with CFD provides a framework for analyzing both near wall and bulk flow parameters. Near wall parameters such as low time-averaged WSS (TAWSS) and high oscillatory shear index have been shown to correlate with plaque formation [8], though more recently other multidirectional flow metrics have gained increasing interest [5,9]. One of these parameters, namely transWSS, showed superior correspondence with the presence of lesions around rabbit intercostal artery ostia compared to TAWSS [5]; however, its significance in humans has yet to be established. These aforementioned metrics are based on the assumption that the preferred alignment of endothelial cells is with the cycle averaged wall shear stress vector. Other multidirectional WSS metrics have been proposed where the preferred alignment of endothelial cells is along the axis of the vessel of interest [10,11].

Flow in the aortic arch is helical by nature and these spiral flow patterns have been suggested to provide an atheroprotective role by reducing luminal surface low-density lipoprotein concentrations [12]. Early work qualitatively identified helical flow patterns [13] though more recently several metrics have been developed to

quantify the amount of helicity in the flow field [7,14,15]. The shape of the aortic arch is believed to play a role in the development of helical flows; however, quantifying aortic arch shape remains a challenge. The use of traditional metrics such as tapering, torsion, and curvature to describe geometry may not fully capture the complex relationship between geometry and hemodynamics. For instance, a local bulging may significantly influence the hemodynamics but these simple metrics do not account for this. A more appropriate approach to examining this relationship may be through statistical shape modeling [16]. This approach provides a statistical platform to identify the extent to which an individual geometry differs from the population average.

In this work, we investigated intraspecies variations in the hemodynamics of the rabbit aortic arch. New Zealand White rabbits undergoing carotid ligation surgery were imaged using 4D PC-MRI and flow was extracted at all arteries of interest. The anatomical images are used to build 3D models of the arch and the measured flow profiles are applied as boundary conditions for CFD analysis. Using this approach, we aim to (i) determine rabbit aortic arch vessel flow rates from 4D PC-MRI, (ii) determine regions of disturbed near wall hemodynamics in the rabbit arch, and (iii) investigate the potential of statistical shape analysis in understanding the role of geometry in the bulk hemodynamics of the vessel.

Methods

Partial Ligation Model. All animal procedures were approved by the Emory University Institutional Animal Care and Use Committee (IACUC). Twelve New Zealand White rabbits on a high fat diet (TD0.6179—regular rabbit diet plus 6% peanut oil and 0.5% cholesterol, Teklad, Madison, WI) underwent either partial left carotid ligation ($n = 8$) or a sham procedure ($n = 4$) (Table 1). Partial carotid ligation is a surgical technique that reduces blood flow to the common carotid artery on the ligated side as previously described in mice [17]. After exposing the carotid bifurcation in anesthetized rabbits under sterile conditions, two silk sutures were used to completely ligate all the branches of the left common carotid artery except one small branch. This resulted in decreased flow to the left common carotid artery and an accompanying decrease in the left common carotid artery diameter. The sham procedure involved exposing the arteries and applying the sutures but not proceeding with ligation, leaving the sutures in place loosely with no effect on carotid flow.

Magnetic Resonance Imaging. A first cohort of rabbits underwent cardiovascular magnetic resonance (CMR) imaging 13 weeks (rabbits 1–5) after surgery, while a second cohort was imaged 6 weeks (rabbits 6–12) after surgery. One hour prior to imaging, anesthesia was induced using ketamine (10 mg/kg) and xylazine (3 mg/kg) intramuscular injection and maintained on

Table 1 Rabbit characteristics at time of imaging

Subject	Age (months)	Weight (kg)	Aortic diameter (mm)	Reynolds no.	Heart rate (bpm)	Ligation
1	12.95	3.55	8.09	193	185	Y
2	12.95	3.35	6.9	287	185	Y
3	12.95	3.1	6.71	262	153	N
4	12.95	3.2	7.05	260	185	N
5	12.95	4	8	451	167	Y
6	5.8	3.6	6.67	292	228	Y
7	5.8	3.5	6.91	239	253	Y
8	5.8	3.3	8	374	165	Y
9	5.8	3.6	7.32	414	215	Y
10	5.8	3.5	6.99	379	178	N
11	5.8	3.6	9.47	307	184	Y
12	5.8	3.2	6.92	351	169	N

Fig. 1 Example of three phase images of the aortic arch (left). Approximate locations from where flow rates are determined by integrating measured velocity over cross-sectional area (right).

1.5–1.8% inhaled isoflurane throughout MRI. Heart rate, respiratory rate, rectal temperature, and blood oxygen saturation were continuously monitored. Electrocardiogram leads were placed on the rabbit before it was positioned feet first and supine in the MRI scanner. MR data were acquired on a Siemens Magnetom Prisma 3 Tesla whole body MRI scanner (Trio, version VB-17, Siemens Medical Solution, Erlangen, Germany) using a flexible 18 channel body coil. Cardiac and respiratory gating were used to obtain consistent and low artifact MR images of periodically moving tissue. A 2D, multiple slice, TurboFLASH MRA sequence was performed to obtain the rabbit vascular anatomy. Following the anatomical data acquisition, a three-dimensional, electrocardiogram-gated cine phase-contrast CMR with three-directional velocity-encoding sequence was performed (Fig. 1). This sequence is more commonly referred to as 4D PC-MRI [18]. A velocity encoding of 120 cm/s was used to determine the velocity field. MR study parameters are summarized in Table 2.

Three-Dimensional Reconstruction. The acquired anatomical images were used to construct 3D models of the rabbit arches. A semi-automatic level set approach was used for image segmentation using the MATLAB-based program (Segment 2.0, Medviso, Lund, Sweden) [19,20]. All side-branches of the aortic arch were included. Due to the large slice thickness with respect to the vessel diameter in most cases, it was not possible to accurately reconstruct secondary supra-aortic branches such as the right subclavian artery or left vertebral artery as the imaging plane aligned with these vessels. Hence, as is often done in studies of aortic arch flow, these secondary branches were ignored [7,10,15]. A stereolithography (stl) file of the reconstructed geometry was generated using the marching cubes algorithm, and subsequently, Laplacian smoothing was applied to the surface mesh.

Boundary Conditions. Inflow (ascending aorta) and outflows (brachiocephalic, left carotid, left subclavian, and descending

aorta) were determined by integrating the through plane velocity component over the cross-sectional area (Fig. 1). In several cases, the left carotid ostia originated from the brachiocephalic artery. When this occurred, flow entering the brachiocephalic artery was measured above where the left carotid artery departed the brachiocephalic artery. A comparison of the inflow and summed outflows found that mass was not conserved. To correct for this, we followed the approach of Trachet et al. to redistribute the flow [21] where the difference in inflow to the summed outflows is redistributed over all outlet boundaries by weighting this according to the outlet boundary diameter.

Computational Fluid Dynamics. Hexahedral meshes ranging between 525,000 and 1,125,000 elements were created using ICEM-CFD (ANSYS, Canonsburg, PA). Grid independence was established for both near wall and bulk flow parameters by incrementally increasing the mesh size until the difference in helicity and TAWSS between successive meshes was less than 5%. The finite volume solver Fluent (ANSYS) was used to solve the Navier–Stokes equations. Blood was assumed to be an incompressible Newtonian fluid with a density of 1050 kg/m³ and a dynamic viscosity of 0.0035 kg/ms. The time-dependent three velocity components measured at the ascending aorta were mapped to the inlet as a velocity inlet boundary condition. The corrected outlet flows were applied as outlet boundary conditions. The arterial walls were assumed to be rigid and a no-slip boundary condition was applied. Unsteady simulations were performed with 300 time-steps per cardiac cycle.

Hemodynamic Analysis. Near wall parameters were assessed by the TAWSS and the cross flow index (CFI). TAWSS is given by

$$\text{TAWSS} = \frac{1}{T} \int_0^T |\tau_w| dt$$

where T is the time period for one cardiac cycle, t is the time, and τ_w is the instantaneous WSS vector. Cross-flow index represents the average over the cardiac cycle of the normalized WSS vector perpendicular to the mean WSS vector [9]. A value of 0 indicates that the flow does not deviate from the time-averaged WSS vector

$$\text{CFI} = \frac{1}{T} \int_0^T \left| \frac{\tau_w}{|\tau_w|} \cdot \left(\mathbf{n} \times \frac{\int_0^T \tau_w dt}{\int_0^T |\tau_w| dt} \right) \right| dt$$

where $\mathbf{n}$ is the normal of the surface wall. Bulk flow hemodynamics were assessed through helicity [22] where the helicity, H , of a flow field with velocity vector $\mathbf{v}$ is given by the integration of the helical density, H_k , over the volumetric domain, D

$$H_k = (\nabla \times \mathbf{v}) \cdot \mathbf{v}$$

$$H = \int_D H_k dV$$

Local normalized helicity (LNH) was determined as

$$\text{LNH} = \frac{(\nabla \times \mathbf{v}) \cdot \mathbf{v}}{|\nabla \times \mathbf{v}| |\mathbf{v}|}$$

where $\nabla \times \mathbf{v}$ is the vorticity. LNH assumes a value between -1 and 1 , where a value of -1 indicates a counter-clockwise helix

Table 2 Magnetic Resonance imaging parameters

	MRA	Phase contrast-MRI
Sequence	2D TurboFLASH	4D Flow CMR
TR/TE/flip angle	110 ms/6 ms/80 deg	32 ms/5.3 ms/15 deg
Slice thickness (mm)	2.5	2
Number of axial slices	22	30–45
Acquisition matrix (pixels)	400 × 400	512 × 352
Field of view (cm × cm)	16 × 16	15 × 10.3
Temporal resolution (ms)	—	30

Fig. 2 Contours of TAWSS (Pa) and CFI for subject 12. Arches are unwrapped about the outer curvature and the inner curvature is denoted by the dashed line. Binary maps are then constructed based on predefined thresholds for abnormal hemodynamics.

and 1 a clockwise helix. Gallo et al. [23] extended the above definitions to generate the following flow descriptors: h_1 —time-averaged helicity, h_2 —helicity intensity, h_3 —the relative balance of counter-clockwise and clockwise helicities, and h_4 —balance of relative rotations (neglects direction of helicity) (See Appendix A for full definitions).

Shape Analysis. Statistical shape analysis was performed using the open source software DEFORMETRIC 2.1 [24]. This software constructs an atlas representing the average rabbit aortic arch from all rabbit geometries using a large deformation diffeomorphic metric mapping framework. The resolution ($\lambda_w = 14.6$ mm) and stiffness ($\lambda_v = 4.6$ mm) parameters, which control how closely deformations of the atlas will match the original geometry, were chosen using the methodology suggested by Bruse et al. [16]. Moment vectors describe the deformation from the atlas to each individual rabbit. Principal component analysis, which is a data reduction technique that aligns data along the main modes of variation of the data, is applied to the moment vectors in order to extract new orthogonal axes. These new axes—the shape modes—are hence defined by the greatest variation of shape in the subjects. For each shape mode, scores, often referred to as the shape vector, are determined by projecting the original moment vectors onto the new axes. Finally, bivariate correlation between the shape vector for each shape mode and parameters of interest such as helicity can be performed.

Statistical Analysis. When relevant, data are presented as mean $\pm$ standard deviation. Geometric metrics including centerline length, tapering, median diameter, volume, mean (signed) torsion, and mean curvature were compared to the bulk flow descriptors using bivariate correlation analysis with Pearson's correlation coefficient r reported. Tapering was defined as the arctangent of the difference in radii between the ascending aorta inlet and the descending aorta outlet divided by the centerline length [3]. Mean curvature and torsion were determined by averaging the curvature and torsion along the centerline at 0.5 mm intervals. Differences between partial ligation and sham ligation flow distributions were evaluated with the student's t -test. All statistical tests were deemed significant at $p < 0.05$. Near wall

parameters were unwrapped and displayed en-face (Fig. 2). This was achieved by projecting the 3D surface values onto an $m \times n$ (length $\times$ circumference) rectangular surface. The length m is given by the length of the centerline divided by 0.5, while n was set to 50 for all rabbits.

Results

Phase Contrast Magnetic Resonance Imaging. Figure 3 illustrates the corrected percentage flow distribution into each vessel. Negligible differences were observed between partial and sham ligation rabbits in the distribution of flow entering the descending aorta ($72.5 \pm 5.3\%$ and $69.5 \pm 2.1\%$, respectively) and the left subclavian artery ($6.4 \pm 1.4\%$ and $6.5 \pm 1.6\%$, respectively). However, the ligation resulted in a significant decrease in left carotid flow compared to the sham ligation rabbits ($3.4 \pm 1.6\%$ and $7.75 \pm 1.7\%$, $p < 0.05$). As a result of the partial ligation, flow entering the brachiocephalic is marginally increased ($17.7 \pm 4.9\%$ versus $16.25 \pm 1.2\%$). A large secondary component (in-plane velocity magnitude) of flow was observed at the inlet. This mean in-plane velocity magnitude during systole was 49.9% of the mean through-plane velocity magnitude.

Effect of Ligation. In order to determine whether the ligation had an impact on aortic arch hemodynamics, rabbit 9 was randomly selected from the population and the flow field was computed using population averaged sham ligation outlet boundary conditions. A second simulation was performed in the same rabbit using population averaged partial ligation outlet boundary conditions (Fig. 3). Negligible differences in aortic arch and descending thoracic aorta TAWSS between different boundary conditions were observed (Fig. 4). A difference of < 1 Pa was observed in 91% of nodes. In a similar manner, 90% of nodes had a difference in CFI of < 0.1 . However, a large difference in TAWSS was observed in the left carotid artery. This can be attributed to the reduced flow entering the vessel when partial ligation outlet boundary conditions are used.

Negligible differences in aortic arch LNH were observed between different boundary conditions (Fig. 5). Both boundary

Fig. 3 Average percentage distribution of ascending aorta flow through each vessel. Error bars indicate standard deviation. * indicates significant difference in vessel flow between partial and sham ligation.

Fig. 4 (a) TAWSS (Pa) of subject 9 with flow distribution using population averaged sham ligation outlet boundary conditions (left) and population averaged partial ligation outlet boundary conditions (right). (b) Difference in subject 9 TAWSS (Pa) between sham and partial ligation boundary conditions.

conditions resulted in the formation of predominantly counter-clockwise helical structures with an equivalent value of $h_3 = -0.16$. Minor differences in the LNH were found in the brachiocephalic and left carotid arteries. As minimal differences in arch hemodynamics were observed going forward, we do not further analyze differences between ligated and nonligated rabbits.

Fig. 5 Local normalized helicity of subject 9 with flow distribution using population averaged sham ligation outlet boundary conditions (left) and population averaged partial ligation outlet boundary conditions (right)

Wall Shear Stress. Time-averaged wall shear stress patterns on the surface of the aortic arch were highly heterogeneous (Fig. 6). However, a region of low TAWSS was consistently observed on the outer curvature of the ascending aorta. In several of the rabbits, low TAWSS was also observed on the inner curvature of the arch.

Cross flow index was found to be high on the outer curvature of the ascending aorta and on the inner curvature of the aortic arch in most cases (Fig. 6). CFI was reduced in the descending aorta with several rabbits illustrating streak-like appearances of CFI.

The colocalization of low TAWSS and high CFI was investigated by determining the % area overlap between both quantities. Low TAWSS was defined as TAWSS less than the 20th percentile of the distribution, while CFI was deemed to be high when CFI exceeded 0.5 (Fig. 3). The surface area along the inner curvature and the outer curvature exposed to low TAWSS and high CFI was calculated where both inner and outer curvature surfaces were defined over a 90 deg arc. The ratio of outer curvature to inner curvature exposure was determined for each case. An average ratio of 3.06 was found, while in 8 of 10 cases a greater surface area in the outer curvature was exposed to low TAWSS and high CFI. Only case 1 showed a substantially greater ratio (2.3) of inner curvature surface area exposed to low TAWSS and high CFI. In almost all cases, low TAWSS and

Fig. 6 Unwrapped surfaces illustrating TAWSS (Pa) (left column), CFI (middle column), and colocalized low TAWSS and high CFI (right column) of each rabbit. Arches are unwrapped about the outer curvature and the inner curvature is denoted by the arrow. Individual rabbit geometries are not shown at the same size scale.

high CFI were colocalized on the outer curvature of the ascending aorta proximal to the brachiocephalic artery (Fig. 6). Subjects 3, 5, 10, and 12 showed regions of colocalization on the aortic arch inner curvature.

Helical Flow. An example of helical flow in the form of LNH is shown in Fig. 7. During systole, structured helical formations develop in the ascending aorta and persist through to the descending aorta. This structured pattern continues during flow deceleration until eventually the structures break down during diastole. Systolic and diastolic helical flow structures for all rabbits can be seen in Appendix B.

Significant correlations were seen between two geometric metrics and helical flow descriptors (Table 3). The centerline

length ($R = -0.65$, $p < 0.05$) was inversely associated with a change in h_4 . The median diameter significantly correlated with an increase in h_4 ($R = 0.61$, $p < 0.05$). There were no other significant correlations between the geometric metrics and the bulk flow descriptors.

In 10 of 12 cases, helicity was predominantly counter-clockwise ($h_3 < 0$) with an average value across all subjects of $h_3 = -0.13$. Figure 8 illustrates the geometry and isosurfaces of time-averaged LNH of rabbits with opposite dominant helical structures. Subject 6 is chiefly characterized by a counter-clockwise helical flow, while in subject 10 helicity is predominantly clockwise.

Statistical Shape Analysis. The atlas generated from the statistical shape analysis was found to be representative of the average geometric features of the rabbit population. Centerline length (52.55 mm), median diameter (2.38 mm), and mean curvature (0.067 mm^{-1}) were all within 5% of the population averages. The first five shape modes derived from the principal component analysis were found to explain 91% (41%, 25%, 12%, 8%, and 5%) of the total shape variance. Correlation coefficients between the shape modes and the helical flow parameters are shown in Table 4. The first shape mode was found to have a significant linear relationship with h_4 ($R = 0.75$, $p < 0.05$). Visually, this shape mode described changes in the aorta length as well as deflections of the ascending aorta away/toward the descending aorta (Fig. 9).

Shape mode 3 showed a nonsignificant correlation with the helicity intensity ($R = 0.56$, $p = 0.058$). Deviations of this shape mode chiefly corresponded to a straightening/bending of the descending aorta (Fig. 10). Shapes with straighter descending aortas had less helicity intensity compared to those with arch configurations with greater bending. Shape mode 5 which primarily described tapering of the geometry was also nonsignificantly correlated with h_2 ($R = 0.53$, $p = 0.156$).

Discussion

The aortic arch in rabbits, as in humans, is the main conduit for blood flow supplying both the head and body. Atherosclerotic lesions are known to form in several locations in this vessel. An understanding of the intraspecies hemodynamics may lead to a better appreciation of how hemodynamics impact lesion formation. With this motivation, we used a combined approach of MRI and CFD to determine the flow field in 12 rabbit subjects. We found low TAWSS and high CFI to colocalize at the ascending aorta and to a lesser extent on the aortic arch inner curvature. A parameter describing the balance of helical flow structures (h_4) was significantly associated with an increase in centerline length and median diameter. Finally, through statistical shape analysis, we found the first shape mode which, corresponded to the largest shape variation in the cohort, showed the greatest correlation with the same parameter.

Prior analysis of rabbit aortic arch hemodynamics has used vascular corrosion casting to reconstruct the 3D surface [3–5,9]. This approach provides a high resolution vessel wall though is subject to shrinkage and does not provide any information pertaining to the flow [25]. To overcome the lack of flow data, several authors have used ultrasonic flowmeter measurements from Barakat et al. [26] who found that 14.7% of flow entered the brachiocephalic artery (including left and right carotid arteries) and 7.1% to the left subclavian artery. The MRI-based approach adopted in this work allowed for the simultaneous gathering of flow and anatomical data albeit at a reduced resolution compared to casting. Using PC-MRI to determine the percentage flow distribution, we found a similar distribution of flow in the left subclavian artery (6.4%) to that reported by Barakat et al. (7.1%). A greater amount of flow was measured in the brachiocephalic artery (17.7%) compared

Fig. 7 Isosurfaces of LNH at five different stages of the cardiac cycle. LNH = 0.8 indicates clockwise helices and LNH = -0.8 indicates counter-clockwise helices.

Table 3 Geometric metrics of individual rabbit geometries (* denotes significant correlation with h_4)

Subject	Centerline length (mm)*	Taper (deg)	Median diameter (mm)*	Volume (mm ³)	Mean torsion (mm ⁻¹)	Mean curvature (mm ⁻¹)
1	52.55	7.04	2.93	2164	-0.01	0.064
2	56.17	6.2	2.19	1452	0.23	0.064
3	43.51	8.32	2.07	1192	0.15	0.079
4	55.09	6.59	2.36	1296	-0.27	0.066
5	51.01	7.78	2.55	1542	0.06	0.073
6	49.97	6.5	2.31	1187	-0.12	0.068
7	47.72	6.8	2.43	1331	-0.14	0.069
8	63.57	6.56	2.36	2030	0.08	0.059
9	54.4	6.93	2.37	1393	0.08	0.063
10	49.63	7.16	2.33	1167	-0.1	0.067
11	65.35	7.84	2.4	2279	0.08	0.055
12	62.54	5.2	2.32	1727	-0.17	0.06
Mean $\pm$ stdev	54.29 $\pm$ 6.7	6.91 $\pm$ 0.82	2.39 $\pm$ 0.2	1443 $\pm$ 598	-0.01 $\pm$ 0.15	0.065 $\pm$ 0.006

Fig. 8 Isosurfaces (top row) of LNH and geometry (bottom row) for rabbits with dominant clockwise ($h_3 > 0$) and counter-clockwise helical flow ($h_3 < 0$)

Table 4 Correlation coefficients between helical flow parameters and shape modes. p values are shown in brackets

	Shape mode 1	Shape mode 2	Shape mode 3	Shape mode 4	Shape mode 5
h_1	-0.355 ($p = 0.25$)	-0.232 ($p = 0.46$)	-0.362 ($p = 0.24$)	-0.282 ($p = 0.37$)	-0.531 ($p = 0.075$)
h_2	0.282 ($p = 0.37$)	0.024 ($p = 0.93$)	0.559 ($p = 0.058$)	0.205 ($p = 0.52$)	0.436 ($p = 0.156$)
h_3	-0.442 ($p = 0.149$)	-0.357 ($p = 0.254$)	0.165 ($p = 0.608$)	-0.076 ($p = 0.812$)	-0.238 ($p = 0.455$)
h_4	0.752 ($p = 0.0048$)	0.145 ($p = 0.652$)	-0.459 ($p = 0.133$)	-0.088 ($p = 0.784$)	0.085 ($p = 0.791$)

Fig. 9 (a) Relationship between shape mode 1 and h_4 . (b) Change in shape of the atlas toward ± 2 standard deviations of the shape mode.

to that of Barakat et al. (14.7%). This difference is actually larger than on first inspection given that in this work we treat the left carotid as a separate measurement. Given the different methods used in both studies to collect these measurements, it is difficult to explain the differences. One possibility is errors in the right carotid and right subclavian measurements of Barakat et al., as these are estimated due to the measurement technique causing occlusion of these vessels. In this study, errors may still arise due to an incorrect reportioning of the flow to each vessel.

It has been hypothesized that the aortic arch shape naturally arises in order to promote helical flow [12]. The curvature and twist of the vessel has been shown to support vortex formation [9] and these flow patterns have been shown to reduce low-density lipoprotein deposition on the luminal surface [12]. Despite this hypothesis, the impact of aortic arch geometry on helicity has not been extensively characterized. We assessed the geometry first using traditional metrics and second using a statistical shape analysis framework. Large centerline length, representative of a geometry including greater length of the descending aorta, was negatively correlated with h_4 . As h_4 goes

Fig. 10 (a) Relationship between shape mode 3 and h_2 . (b) Change in shape of the atlas toward ± 2 standard deviations of the shape mode.

to zero, an equal amount of clockwise and counter-clockwise helical structures is found in the flow field. The correlation with centerline length may be attributed to the development of counter-rotating helical structures balancing each other out as the flow continues into the descending aorta. The statistical shape analysis revealed that the first shape mode, which primarily appeared to describe a change in centerline length, was also associated with increased values of h_4 . However, on closer inspection, we found that this shape mode was not significantly associated with either centerline length ($R = -0.54$, $p = 0.067$) or median diameter ($R = 0.34$, $p = 0.276$). While the correlation with the centerline length approaches statistical significance, there may be other aspects of this shape feature, such as the deflections of the ascending aorta which contribute to the flow helicity. This could be investigated in more detail with idealized models. Counter-clockwise helical flow was found to be the dominant direction of helicity (mean $h_3 = -0.13$). However, this was not associated with any traditional geometric metrics or any of the shape modes from the statistical shape analysis. Tapering of the aorta through the descending aorta has previously been shown to maintain helical flow [12]. Interestingly, the fifth shape mode was significantly correlated with vessel tapering ($R = 0.68$, $p < 0.05$) and this shape mode also showed

a positive trend with the helicity intensity (h_2). However, this mode did not reach statistical significance ($R = -0.53$, $p = 0.075$). A further nonsignificant positive correlation was found between the third shape mode and the helicity intensity. The helical intensity was, however, correlated with an increasing Reynolds number ($R = 0.85$, $p < 0.05$). At higher Reynolds numbers in curved tubes, the Dean number increases resulting in increased secondary flows. Comparison with prior studies of human aortic arch helicity is challenging due to the generally qualitative nature of these studies and the large variability among individual humans. However, a right-handed helix is commonly reported in the region of the upper arch [13,27], while bi-helical structures have been shown to exist in the ascending aorta [28,29]. These counter-rotating bi-helical structures were commonly seen in the ascending aorta of the rabbits but generally the disappearance of the left-handed helical flow in the arch was not observed. In diastole, a breakdown of coherent helical flow structures was observed as has previously been described in humans [27].

Low TAWSS has been linked with the formation of lesions in humans and animals. However, the correlation between lesions and low TAWSS has been criticized as being mostly supported by qualitative or selective evidence [30]. A newly developed metric, transWSS, motivated by the alignment of endothelial cells to flow, was found to correlate with lesion formation in rabbit intercostal arteries. Here, we investigated the normalized value of transWSS—CFI—and we identified regions where low TAWSS and high CFI were colocalized. While values of $WSS < 0.5$ Pa are considered pro-atherogenic in humans, thresholds for low WSS have not been established in rabbits. For this reason, we selected a threshold of the 20th percentile of the WSS distribution as a potential cut-off for disturbed WSS. For high CFI, we selected a threshold of $CFI > 0.5$. These chosen thresholds are somewhat arbitrary and future in vitro studies should be performed to establish values at which rabbit endothelial cells become pro-atherogenic. The outer curvature of the ascending aorta was the only location consistently found to have both variables colocalized. The inner curvature, an often cited location of lesion formation, was only found to have colocalized high CFI and low TAWSS in subjects 3, 5, 10, and 12.

Limitations. Ideally, all rabbits would have been imaged at the same time after surgery, however, due to logistical challenges, this was not possible. This time difference (6 versus 13 weeks) was not believed to influence the flow or anatomy in the ligated subjects as (based on the investigators experience) flow and anatomical changes occur within the first 2 weeks of the procedure and remain constant after this. Due to the high fat diet, extensive fatty deposits are present in the arch. These may have had an impact on the geometry and resulting hemodynamics from what would be expected in an arch free of disease. Compliance and motion of the aorta was neglected. Prior studies have indicated that flow patterns in the aorta are affected not only by the geometry but also by the vessel deformation and the exclusion of aortic motion results in inferior agreement with in vivo measurements [27].

The geometry derived from MRI does not preserve small-scale features as well as cast models. For example, Vincent et al. [4] have previously found the ductus arteriosus scar, a feature beyond the resolution of MRI, to produce significant deviations in vessel WSS. Further, the low number of cross sections used to reconstruct the geometries can result in a less accurate aortic arch inner curvature surface. Lower accuracy aortic arch surfaces have previously been shown in mice to result in altered shear stress patterns on the arch inner curvature [31]. Since mass was not conserved when summing the outflows, a redistribution scheme was employed. This redistribution scheme may result in errors in the splitting of flow

between the branches. The measurement error is most likely attributable to the small size of the supra-aortic branches. An alternative strategy of measuring the flow along the aortic arch upstream and downstream of the supra-aortic branches has been shown to alleviate this problem [32]. Finally, the small number of subjects used to construct the atlas may not cover the full range of rabbit aortic arch geometries.

Conclusion

Intraspecies differences exist in the near wall and bulk flow hemodynamics of the rabbit aortic arch. High CFI and low TAWSS were colocalized predominantly in the ascending aorta and to a lesser extent on the inner curvature of the aortic arch. Helical flow intensity is primarily determined by the Reynolds number. A stronger correlation was found between the helical flow parameters and the first shape mode derived from statistical shape analysis than traditional shape metrics illustrating its potential for.

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Appendix A

Gallo et al. introduced quantitative descriptors of helical flow [23]. The helicity density H_k was used to compute the following quantities:

$$h_1 = \frac{1}{T} \frac{1}{V} \int_T \int_V H_k dV dT$$

$$h_2 = \frac{1}{T} \frac{1}{V} \int_T \int_V |H_k| dV dT$$

$$h_3 = \frac{h_1}{h_2}$$

$$h_4 = \frac{|h_1|}{h_2}$$

where h_1 is the time-averaged helicity, h_2 is the helicity intensity, h_3 is a nondimensional quantity ranging between -1 and 1 , taking a value of -1 when only counter-clockwise helical structures are present and 1 when clockwise helical structures dominate, and h_4 determines the balance between counter-rotating helical structures neglecting the major direction of rotation.

Appendix B

The LNH during systole and diastole for all rabbits is shown in Fig. 11

Fig. 11 Local normalized helicity of all rabbits during peak systole (S) and diastole (D)

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