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Variations in the branching pattern of the aortic arch: an African perspective

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SUMMARY

Variation in the branches of the aortic arch is higher in individuals of African descent. However, published studies are few. The aims were to document variations in the branching pattern of the aortic arch in a South African sample, determine whether these variants are more common than other populations, and determine whether there are any significant differences in the prevalence of variation between males and females. The aortic arch and main branches were dissected in 733 cadavers. All branching patterns were documented and classified as types. Chi-Square tests were used to determine whether there were any significant differences in prevalence of variation between males and females. The diameters of the main branches were measured and compared between sexes.

The standard branching pattern was present in 65% of individuals, similar to that reported for other African studies, but lower than other studies from around the world. Variations were more prevalent in males than in females (p = 0.025), while only the diameter of the left vertebral artery, when arising from the arch was significantly larger in females, with no differences between sexes for the other vessel diameters. The results of this study support the hypothesis that variations in the branching pattern of the aortic arch are more common in African individuals. These individuals may be at increased risk of associated although rare, clinical symptoms or iatrogenic injury.

Key words: Aortic arch – Arch of aorta – Branching pattern – Anatomical variation – Aberrant right subclavian artery – Arteria lusoria – Vertebral artery

INTRODUCTION

The aortic arch gives rise to three arteries known as the brachiocephalic trunk (BCT), the left common carotid (LCC) and the left subclavian artery (LSA) (Popieluszko et al., 2017). Several variations in the number and order of these branches have been described (Popieluszko et al., 2017). The most common is a common origin of the BCT and the LCC (Reinshagen et al., 2014), occasionally termed the “bovine trunk” (Layton et al., 2006), with a reported prevalence of 13.6% (Popieluszko et al., 2017).

The left vertebral artery (LV), which usually arises from the left subclavian artery, has been observed as originating directly from the aortic arch in 2.9% (Popieluszko et al., 2017), usually distal to the LSA (Ergun et al., 2013).

The right subclavian artery (RSA) may originate from the aortic arch, or descending aorta, instead of from the brachiocephalic trunk. This “aberrant right subclavian artery” (ARSA) or “arteria lusoria” may be present simultaneously with a common origin for the common carotid arteries in some cases (Ergun et al., 2013). The prevalence of ARSA worldwide has been reported as 0.7%, and the

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presence of a common origin of the carotid arteries as 0.3% (Popieluszko et al., 2017).

While most variations of the aortic arch are clinically "silent", some may have clinical consequences (Celikyay et al., 2013). A common origin of the BCT and the LCC contains three of the four arteries supplying the brain (Azakie et al., 1999). Occlusion of this common trunk by atherosclerotic plaques could result in disrupted blood flow to the brain, with stroke as a possible consequence, particularly if the LV is also occluded.

The origin of the LV from the aortic arch has an increased risk of dissection, resulting in decreased blood flow to the brain and subsequent stroke (Komiyana et al., 2001).

Compression of the oesophagus and trachea, associated with pain, difficulty in swallowing, and respiratory symptoms, may be caused by an ARSA (Mahmodlou et al., 2014).

Variant branches may be susceptible to iatrogenic injury during surgical procedures, e.g. an ARSA may be injured during oesophageal surgery (Mahmodlou et al., 2014). In addition, variation may increase the technical difficulty of carotid artery stent placement in cases of a common origin of the BCT and the LCC (Faggioli et al., 2007).

Geographical differences in the prevalence of variant patterns have been reported (De Garis et al., 1933; Celikyay et al., 2013; Reinshagen et al., 2014; Karacan et al., 2014; Popieluszko et al., 2017). It has been suggested that variation is more prevalent in individuals of African descent (Williams et al., 1932; De Garis et al., 1933; Williams et al., 1935; Popieluszko et al., 2017).

However, studies from Africa are fewer than those from the other main continents, which is a limitation when the prevalence of variation is compared between studies. The aims of this study were to: a) test the hypothesis that variation is more prevalent in African individuals by determining the prevalence of variation in the aortic arch; b) measure the diameter of the vessels arising from the aortic arch, and c) determine whether there are any differences in the prevalence of variation and vessel diameter between males and females.

MATERIALS AND METHODS

Study Design

During 1962-2018, a cross-sectional study of 733 formalin-fixed embalmed adult bodies was undertaken at a South African university. The sample comprised of all bodies that were being dissected by second year medical students. Exclusion criteria included any surgical interventions: e.g. total arch replacement, aneurysm repair or damage from previous dissection. This study was performed and reported in compliance with Anatomical QUality Assurance (AQUA) Checklist (Supplement 1).

Dissection

Examination of the aortic arch was performed by four anatomists: one in 1962-1972 and two in 2008-2018 at the University of Cape Town, and fifty-seven of the arches were dissected at a second South African institution by a fourth individual in 1971. The thorax and neck were dissected with a standard dissection kit and technique. The anterior ribcage was removed after being cut on the lateral sides by an oscillating saw (blade cut edge of 94.0 mm, Stryker, USA), after which the pericardium was resected from the heart with a scalpel (size 10 blade), exposing the aortic arch. The fascia covering the aortic arch and its main arteries were removed, allowing the branches to be visualized. The BCT was followed until its division into the right common carotid (RCC) and right subclavian arteries. The LCC, LSA and LV were also exposed.

Data collection

Primary and secondary outcomes

The primary outcomes investigated in this study were a) the variant types of aortic arch branching patterns and, b) diameter of the vessels arising from the arch. Secondary outcomes included determining any statistically significant differences in the primary outcomes between males and females.

The branching pattern was documented for each arch, and any variant patterns were photographed (Canon EOS 70D) and recorded in Microsoft Word. The prevalence of each variant pattern was determined and expressed as a percentage in Microsoft Excel.

Classification of variant patterns was in accordance with Popieluszko et al. (2017), where possible. As the description of variant types has been recently published, the initial data collected in 1962-1972 could not be classified. The data collected in 2008-2018 was classified into types, which are described as follows (Fig. 1):

Type 1: standard pattern of three branches (BCT, LCC, LSA).

Type 2: common origin of the BCT and LCC.

Type 3: origin of the LV from the aortic arch.

Type 4: combination of Type 2 and Type 3, namely a common origin of the BCT and LCC in conjunction with the LV originating from the arch.

Type 5: absence of the BCT, instead a common trunk for the left and right common carotid arteries, with the RSA originating directly from the arch.

Type 6: ARSA.

Type 7: right-sided aortic arch.

Measurements

The internal diameters of the BCT, LCC, LSA and the LV, in cases where it originated from the arch, were measured with an electronic digital Vernier caliper (ORIGIN 0-150 mm, China) in a subset of the sample. In addition, the width and height of any common trunk for the BCT and LCC was measured. The height was measured from the point of origin on the arch until the point at which the common trunk bifurcated into BCT and LCC. Each measurement was taken three times and the

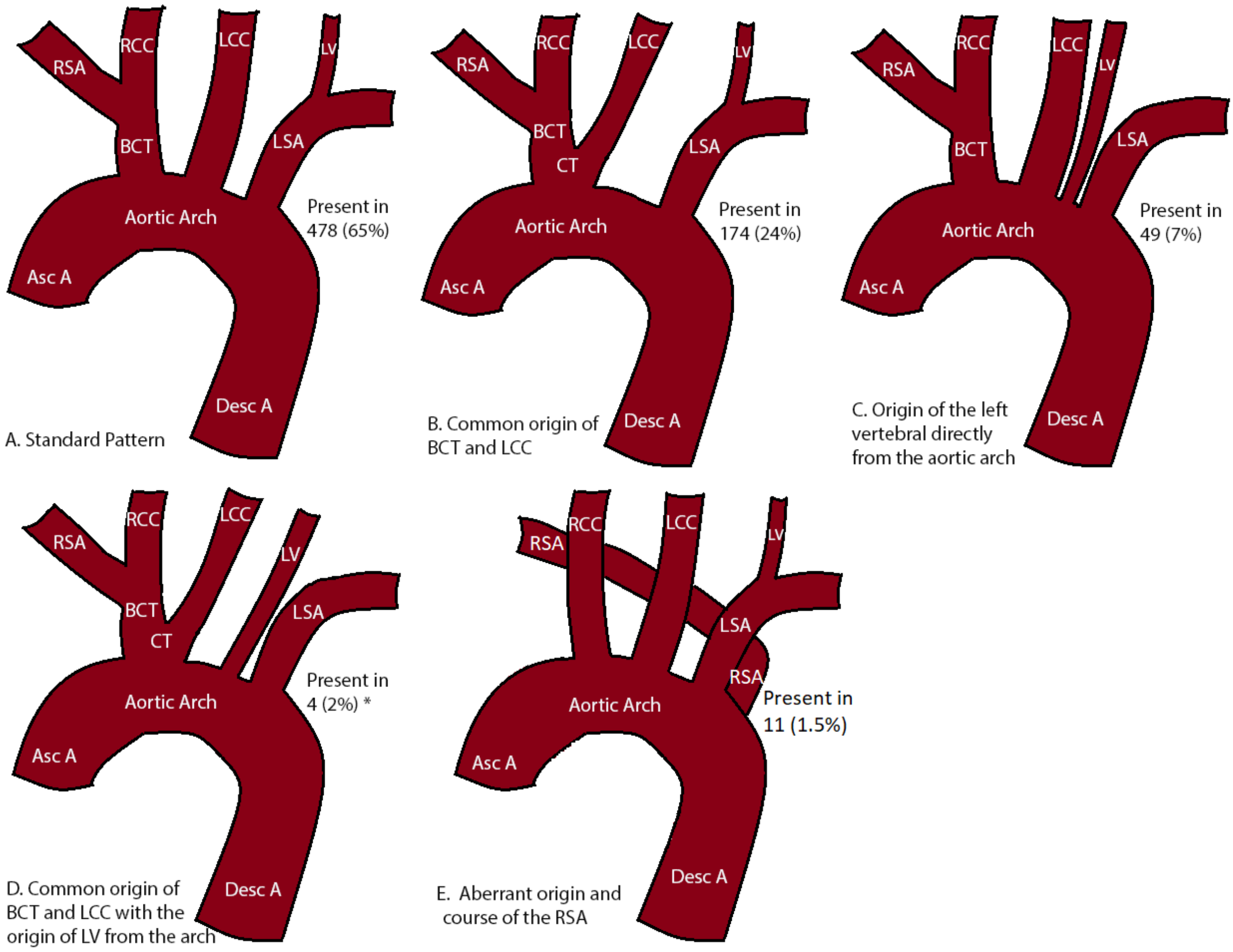

Fig 1. The most common branching patterns (**A-E**) observed in the sample, with the prevalence reported for each pattern. *Pattern D was only recorded for the subset 2008-2018, in a total sample of 197. Abbreviations: Asc A - ascending aorta, BCT - brachiocephalic trunk, RSA - right subclavian artery, RCC - right common carotid artery, LCC - left common carotid artery, LSA - left subclavian artery, LV - left vertebral artery, Desc A - descending aorta, CT - common trunk.

average recorded in Microsoft Excel for analysis.

Statistical analysis

Statistical analyses were performed in IBM SPSS® Version 24.0 (Armonk, New York, United States).

Categorical data were represented as number (n) and percentage (%). Numerical data were represented as summary statistics, namely mean ± standard deviation (SD) for normally distributed data, or median and inter-quartile range for non-normally distributed data. Normal distribution of the numerical data was determined with the Shapiro-Wilk test.

Significant differences in the prevalence of variant types were investigated between males and females using Chi-square or Fischer-Exact tests. Differences in the diameter of the main arteries between sexes were investigated by means of unpaired Student t-tests.

A p-value of less than 0.05 was considered to be significant for all statistical analyses.

Ethical compliance

The body donation program at the University of Cape Town complies with the International Federation of Associations of Anatomists (IFAA)'s "Recommendations of good practice for the donation and study of human bodies and tissues for anatomical examination". In accordance with our institutional review board, as written informed consent was obtained from body donors for teaching and research purposes, it was not necessary to seek ethical approval from our Human Research Ethics Committee. Consent for the use of unclaimed individuals was granted by the Department of Health, Western Cape Government, South Africa.

RESULTS

Subject Characteristics

The sample comprised of 516 males and 217 females. Information about age at death was only available for subjects in 2008-2018, which ranged from 20-102 years, with a mean age of 64.3 ± 20.8 years. All individuals had died from natural causes. No medical history was available for any of the

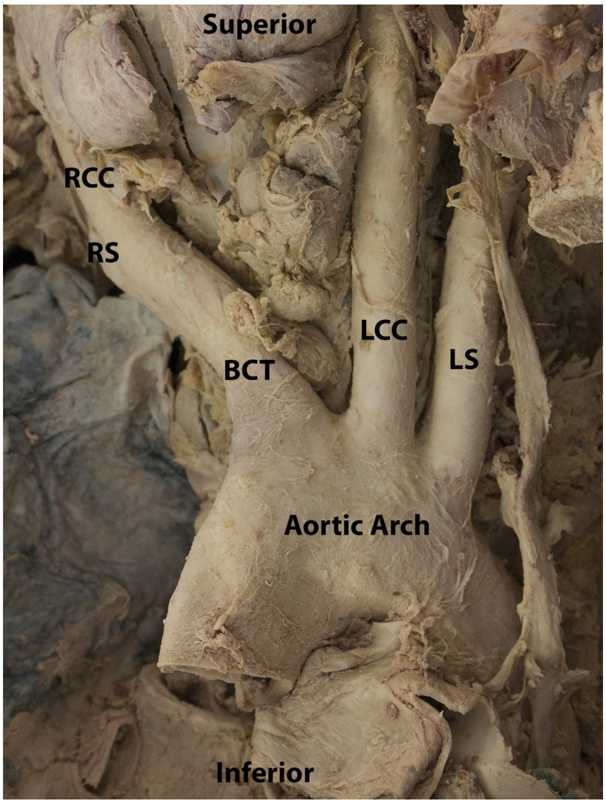

Fig 2. Standard branching pattern of the aortic arch, also known as Type 1. The brachiocephalic trunk (BCT) is the first branch, giving rise to the right subclavian (RS) and right common carotid (RCC), the second branch is the left common carotid (LCC), and the left subclavian (LS) the third branch.

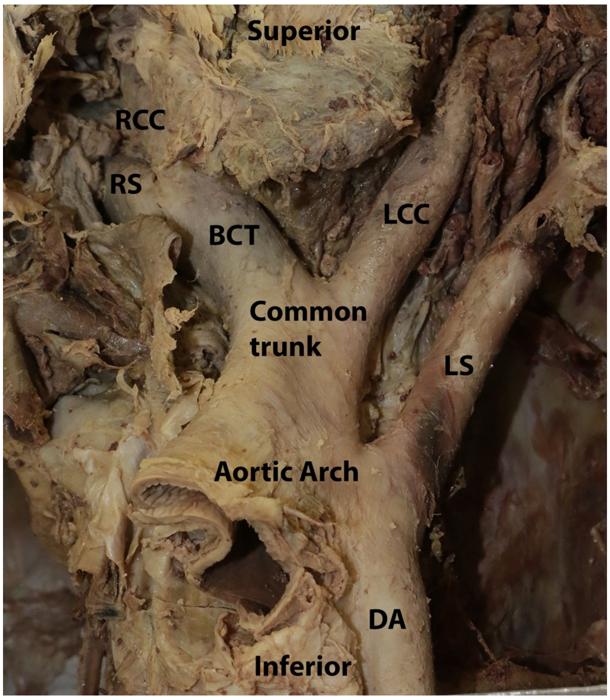

Fig 3. A common trunk for the brachiocephalic trunk (BCT) and the left common carotid artery (LCC), also known as Type 2. Abbreviations: RS - right subclavian, RCC - right common carotid, LS - left subclavian, DA - descending aorta.

cadavers.

Arch patterns were determined in all 733 bodies, while a subset from 2017 was available for measurement of vessel diameters. The diameter of LV was measured in seven cases in which it originated from the arch. The RSA, LCC and LSA were measured in 37 individuals, of which 23 were male and 14 were female. In two male individuals, the BCT had been damaged by prior dissection. These individuals were thus excluded from the analysis. The height and width of a common origin for the BCT and LCC was measured in eight individuals. The diameter of the ARSA could not be determined, as this pattern was not present in the subset available for measurement. Individuals with ARSA had already been cremated.

Prevalence of the branching pattern types

Type 1 pattern was present in 478 out of the 733 individuals (65.2%) (Fig. 1A and Fig. 2). Variant patterns were thus present in 35% of the sample.

The variant branching patterns are summarized in Table 1 and Figs. 1, 3-4. The ARSA was present in 11 (1.5%) of individuals (Figure 1E and Figure 6), for whom no demographic information was available. The most recent case of ARSA has been published as a case report (Keet and Gunston, 2018).

The following rare variants were each observed in one individual (0.14%): right sided arch (published as a case report; Jarvis, 1966), double aortic arch (published as a case report; Jarvis and Shofield, 1967); common origin for the LCC and the LSA; and origin of the LV from the LCC. A common origin of the left and right common carotid arteries with the RSA originating as the first branch of the arch (Type 5) was not observed. In 17 individuals, a more proximal origin than usual of the LV from the first part of the left subclavian artery was noted. This position is not discussed further as the origin of the vertebral artery is nevertheless, from the first part of the subclavian artery as defined in anatomical texts.

Furthermore, information obtained from 1962-1972 could not be classified into pattern types, as only the common origin of the BCT and the LCC, or the LV originating from the arch was documented. There were no descriptions of these two variants occurring together (i.e. Type 4), nor was the absence of this type commented on. Therefore, we cannot classify Types 2, 3 and 4 for the entire sample, and instead we limited this classification to the subset from 2008-2018 (n=197) (Table 2). Type 4 was present in 2% (Fig. 1D and Fig. 5).

Each of the variant patterns were statistically more prevalent in males than in females ($p = 0.025$) (Table 1).

Diameter of main aortic arch branches

All of the measurements were normally distributed with the exception of the diameter of the RSA in males. Only the diameter of the LV from the arch was significantly larger in females than in males (Table 3). None of the other measurements were

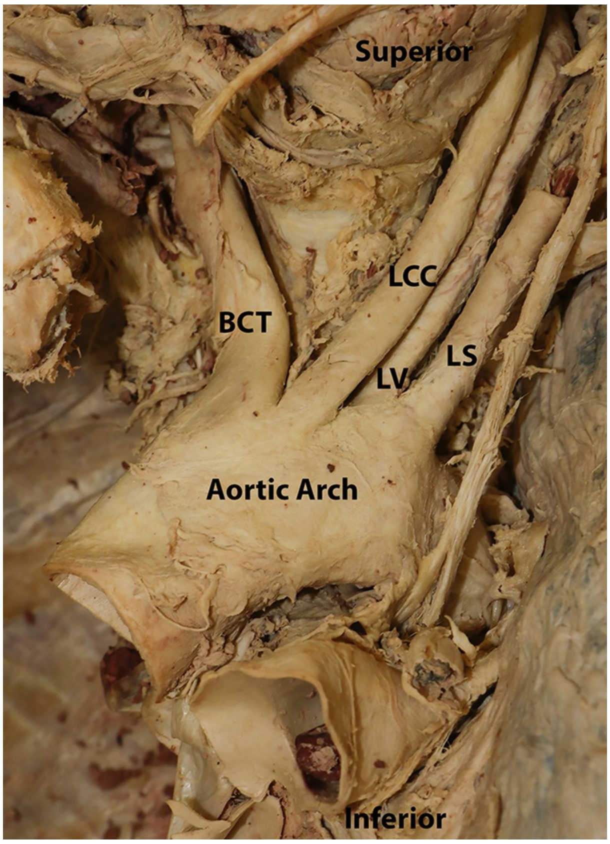

Fig 4. Origin of the left vertebral artery (LV) directly from the aortic arch, also known as Type 3. Abbreviations: BCT - brachiocephalic trunk, LCC - left common carotid, LS - left subclavian.

significantly different between sexes.

DISCUSSION

The standard branching pattern (Type 1) of the aortic arch was present in 65.2%, which is similar to the frequency reported for other African samples and lower than that of other countries (Table 4) (Popieluszko et al., 2017). Ogeng'o et al. (2010) and Makhanya et al. (2004) reported a similar prevalence of the standard pattern to our study (67.3% and 65% respectively). However, Satyapal et al. (2003) observed the standard pattern in 94.7%. It is unclear why the prevalence of variation observed by Satyapal et al. (2003) was lower than the other studies. Dissection studies, such as this study and Ogeng'o et al. (2010) are able to directly visualize the branches of the aorta. Satyapal et al. (2003) and Makhanya et al. (2004) both utilized angiography to study the branching pattern of the aortic arch. Angiography, however, may not display arteries that are occluded, which could then be misinterpreted as absent. In addition, larger vessels may obscure smaller ones, such as the origin of the LV from the aortic arch (Celikyay et al., 2013; Karacan et al., 2014). Angiography does not display other soft tissues, therefore it is more difficult to determine relationships between vessels and adjacent anatomical structures, such as the relationship between an ARSA and the trachea and oesophagus (Celikyay et al., 2013). Multidetector computed tomography (MDCT) may be utilized as an alternative to angiography, as it is noninvasive and high-quality three -dimensional images can be obtained, revealing the relationship of blood vessels to surrounding structures (Celikyay et al., 2013).

The prevalence of a common origin of the BCT and LCC was 23.7%. This is lower than the 26.8% reported for other African studies, although higher than the prevalence reported for international studies (Popieluszko et al., 2017). Origin of the LV directly from the aortic arch was observed in 6.7%, higher than the average prevalence reported for Africa (2.3%) and the rest of the world (Table 4) (Popieluszko et al., 2017). However, this is similar to the 5% reported by Vorster et al. (1998) in a South African sample.

Type 4 was observed in 2% of the subset, higher than the prevalence reported for other studies from Africa (1.2%) and the rest of the world (Table 4). The presence of an ARSA (Type 6) was noted in 1.5%. This is similar to the prevalence reported by Popieluszko et al. (2017) for African studies (1.4%), while higher than that reported for other countries (Table 4). A right-sided arch (Type 7) was present in one individual (0.14%), lower than the 0.3% reported for other African samples

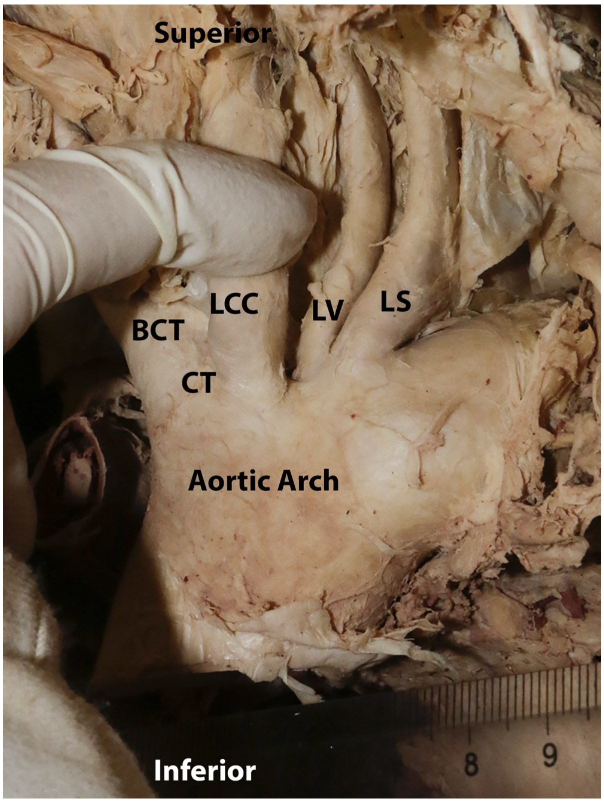

Fig 5. A common trunk (CT) of the brachiocephalic trunk (BCT) and left common carotid (LCC) in combination with the left vertebral artery (LV) originating from the aortic arch, also known as Type 4. Abbreviation: LS - left subclavian.

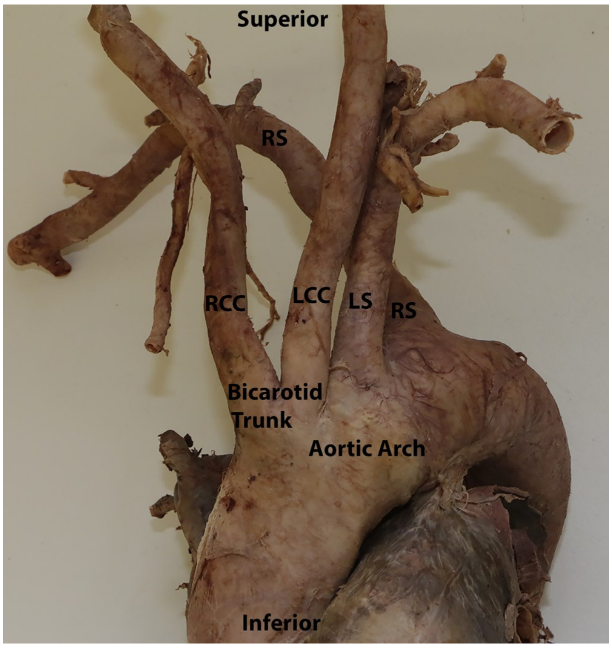

Fig 6. Aberrant right subclavian artery originating as the last branch of the aortic arch, distal to the left subclavian (LS), also known as Type 6. In this particular individual, a bicarotid trunk giving rise to the right common carotid (RCC) and left common carotid (LCC) was present, while the aberrant right subclavian (RS) coursed posterior to the oesophagus and trachea to reach the right upper limb.

(Popieluszko et al., 2017); and a double aortic arch was also present in one individual (0.14%). Satyapal et al. (2013) described two pediatric cases of double aortic arch, while Makhanya et al. (2004) and Ogeng'o et al. (2010) did not report this variant. Although Type 5 pattern was not observed in this study, it has a reported prevalence of 0.3% in African samples (Popieluszko et al., 2017).

All variant patterns were more common in males than in females (p = 0.025), however as the sample consisted predominantly of males, this information should be interpreted with caution. No other African studies have compared the prevalence of variation between sexes (Popieluszko et al., 2017). Some international studies reported no significant differences in the prevalence of variation between males and females (Karacan et al., 2013; Mustafa et al., 2017), while others suggested that certain branching patterns occur more frequently in either males or females (Molz, 1976; Natsis et al., 2009; Piyavisetpat et al., 2011; Boyaci et al., 2015).

There were no significant differences in the diameter of arteries between males and females, with the exception of the LV arising from the arch, which was larger in females. Although a previous South African study has determined the validity of measurements from cadaveric material when compared with CT scans, these authors do not report the diameters of the vessels (Schoeman et al., 2018). Thus, we were unable to compare our measurements with another African study. Knowledge of arterial diameter is relevant in endovascular procedures and transcatheter aortic valve implantation (TAVI) (Valsecchi et al., 2006; Pascual et al., 2017).

The aortic arch and main branches arise from six paired embryological arches (Kondori et al., 2016). The ventral aorta is connected to the paired dorsal aortae via these six arches (Makhanya et al., 2004). The aortic arch originates from the aortic sac, the left fourth arch and a portion of the left dorsal aorta (Mustafa et al., 2017). The right fourth aortic arch, right seventh intersegmental artery and the right dorsal aorta contribute to the RSA. The third arches give rise to the common carotid arteries, with the right arch forming the RCC and the left arch becoming the LCC. The left seventh intersegmental artery forms the LSA (Kondori et al., 2016).

Factors that influence the formation of the aortic arch and its branches may result in variation, such as persistence of a vessel that should regress, or regression of a vessel that typically persists (Yokoyama et al., 2010; Kondori et al., 2016).

The part of the aortic arch in between the respective origins of the BCT and the LCC is formed from the left limb of the aortic sac (Mustafa et al., 2017). If the sac does not divide into left and right limbs, the LCC joins this sac, forming a common origin for this vessel and the BCT.

Alternatively, if the proximal portion of the third arch is absorbed into the right horn of the aortic sac instead of the left horn, this will also result in this variant pattern. Gold et al. (2018) suggested that the presence of this variant vessel could be an independent risk factor for cardioembolic stroke, as patients with this variant have a 50% chance of developing either a right- or left-sided cerebral infarct.

The first part of the vertebral artery usually originates from the seventh intersegmental artery (Lale at al., 2014). Origin of the LV from the aortic arch results when this vessel develops instead from the sixth cervical intersegmental artery together with the persistence of a portion of the dorsal aorta (Lale et al., 2014; Kondori et al., 2016). When the left vertebral arteries arise from the aortic arch, they are often hypoplastic and enter the foramen transversaria at a different level from the C6 vertebrae (Kośla et al., 2014). This must be considered when planning surgical treatment. In addition, vertebral arteries arising from the arch have an increased risk of dissection, which could be caused by altered cerebral vascular hemodynamics or structural defects in the wall of this artery (Berko et al., 2009).

The ARSA results from the degeneration of both the right fourth aortic arch and the proximal part of the right dorsal aorta. Thus, the right seventh cervical intersegmental artery and the distal right dorsal aorta continue as the RSA (Satyapal et al., 2003; Mustafa et al., 2017).

Surgical resection of the ARSA and reattachment to the right side of the aortic arch can alleviate symptoms associated with this variation, such as

Table 1. Prevalence of the most commonly observed branching patterns of the aortic

	Total	Standard Pattern n(%)	Common origin of brachiocephalic trunk and left common carotid n (%)	Left vertebral originating from aortic arch n (%)
Total	733	478 (65.2)	174 (23.7)	49 (6.7)
Males	516	354 (68.6)	111 (21.5)	32 (6.2)
Females	217	124 (57.1)	63 (29.0)	17 (7.8)

Table 2. Prevalence of the most commonly observed variation types in the

	Total	**Type 1 n (%)**	**Type 2 n (%)**	**Type 3 n (%)**	**Type 4 n (%)**
Total	197	124 (63.0)	44 (22.3)	10 (5.1)	4 (2.0)
Males	131	87 (66.0)	29 (22.1)	7 (5.3)	2 (1.5)
Females	66	37 (56.1)	15 (22.7)	3 (4.5)	2 (3.0)

Table 3. Diameter of aortic arch branches

	Mean (SD) (mm)	
Artery	**Males**	**Females**
Brachiocephalic trunk	11.8 (1.6)	10.9 (1.7)
Right subclavian	9.0 (2.5)*	9.2 (1.5)
Right common carotid	7.7 (1.0)	7.3 (1.1)
Left common carotid	7.9 (1.0)	7.6 (0.9)
Left subclavian	9.2 (1.6)	8.5 (1.5)
Left vertebral	3.8 (1.5)	4.3 (0.2)**
Common trunk** *height	10.7 (4.5)	10.9 (2.5)
Common trunk*** width	17.6 (1.3)	17.6 (2.2)

*median (IQR), data not normally distributed, ** Significantly different (p = 0.025), *** for BCT and LCC.

dysphagia and dyspnea (Suzuki et al., 2005; Mahmodlou et al., 2014). When these nonspecific symptoms are reported, the presence of an ARSA should be considered a differential diagnosis (Kaldararova et al., 2017; Jarvis and Shofield, 1967). A patient at our hospital was misdiagnosed with depression after significant weight loss over three years, for which the cause of dysphagia was eventually revealed on CT imaging to be from an ARSA (Rogers et al., 2011).

The presence of an ARSA is higher in disorders such as Down's, DiGeorge, and Edwards' syndromes, and in patients with tetralogy of Fallot (Polguj et al., 2014). This vessel is also associated with the "non-recurrent" course of the inferior laryngeal nerve. Surgeons should be aware of this anatomical variant during thyroid surgery (Pelizzo et al., 2017).

Variation in the branches of the aortic arch may be associated with congenital heart defects. Reinshagen at al. (2014) reported that 98.4% of paediatric patients with a common origin of the BCT and the LCC, which is the most common variant of the aortic arch, had at least one congenital heart defect, including ventricular septal defect, atrial septal defect, and patent ductus arteriosus. Some of these patients also had genetic syndromes such as Down's. It is possible that variant arch anatomy may be a marker of congenital heart disease, although further studies are required.

Knowledge of the possible variants of the aortic arch that may be present have practical implications for surgeons, to avoid iatrogenic injury, for radiographers to avoid misinterpretation of images, and for endovascular surgeons who may be using these vessels as approach pathways or for stent placement (Satyapal et al., 2003, Faggioli et al., 2007; Natsis et al., 2009; Ogeng'o et al., 2010; Ergun et al., 2013; Prada et al., 2016; Rojas et al., 2016; Mustafa et al., 2017). Multidetector computed tomography with contrast provides a clear image of the branching pattern of the aortic arch (Karacan et al., 2014). It is important to know whether variant patterns are more common in a particular geographic group, as these individuals may be at a higher risk of associated congenital heart defects, surgical complications or misdiagnosis. The least variable population with respect to the branches of the aortic arch is suggested to be American Japanese individuals from Hawaii (Nelson and Sparks, 2001). Our study provides further evidence that variant aortic arch branching patterns may be more common in African individuals.

Limitations

The sample was comprised of unequal numbers of males and females, thus the comparison of differences in prevalence of variation between sexes may not reflect the prevalence in the population. Further studies with equal numbers of males and females are required.

Medical histories were not available for the individuals in the sample, and thus no clinical infer-

Table 4. Comparison of the prevalence of variations in the aortic branching pattern observed in the present study with those reported by Popieluszko et al. (2017)

Sample	Standard pattern (%) (95% CI)	Common origin of brachiocephalic trunk and left common carotid (%) (95% CI)	Left vertebral originating from aortic arch (%) (95% CI)	Common origin of brachiocephalic trunk and left common carotid and left vertebral from arch (%) (95% CI)	Aberrant right subclavian artery (%) (95% CI)
Present study	65.2 (61.6-68.7)	23.7 (20.7-27.0)	6.7 (5.0-8.7)	2 (0.6-5.1)	1.5 (0.8-2.7)
Africa	65.8 (51.1-80.2)	26.8 (14.4-41.9)	2.3 (0.0-8.2)	1.2 (0.0-5.7)	1.4 (0.0-6.2)
Europe	82.0 (75.8-85.7)	13.6 (9.4-18.0)	2.3 (0.7-4.5)	0.3 (0.0-1.1)	0.8 (0.0-2.1)
Asia	86.9 (81.5-89.0)	7.4 (4.7-10.3)	3.5 (1.7-5.7)	0.4 (0.0-1.2)	0.5 (0.0-1.7)
North America	78.4 (67.6-84.7)	15.5 (8.6-23.2)	1.9 (0.0-5.1)	0.7 (0.0-2.9)	1.1 (0.0-3.6)
South America	69.5 (48.5-88.2)	24.2 (8.0-45.4)	3.9 (0.0-14.4)	0.8 (0.0-7.0)	0.2 (0.0-4.6)

ences can be made as to whether any of these variations were symptomatic.

The arterial diameters could only be measured in a sample subset, while the ARSA could not be measured. Future imaging studies could address this limitation.

Future studies will be undertaken to quantify the normal origin of the LV from the first part of the subclavian artery, in order to clarify cases where a “proximal” origin may be reported.

CONCLUSION

This study supports the current hypothesis that variation in the branching pattern of the aortic arch is more common in African individuals. Variant patterns were more prevalent in males, and most of the vessel diameters were not significantly different between the sexes. Certain variations may produce dyspnea and dysphagia, which may be difficult to diagnose. Awareness of the prevalence and types of variant patterns is relevant in open and endovascular surgery, as well as medical imaging, and may even be a marker of congenital defects.

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EVIDENCE BASED ANATOMY INTERNATIONAL WORKING GROUP

Anatomical Quality Assurance (AQUA) Checklist

"For improving the quality and reporting of anatomical studies."

Checklist Component	#	Description and Recommendation:	Page Number*
Title			
Title	1	Identify the main objective or key characteristic of the study in the title.	1
Abstract			
Structured Summary	2	Provide a clear and structured summary of the study with emphasis on the aims, methodology, key findings, and conclusions directly supported by study findings.	2
Introduction			
Background / Rationale	3	Provide a rationale for the study including a concise, updated scientific background, appropriately referenced. Identify any relevant knowledge gaps to support the study rationale.	3-4
Objective	4	Indicate clearly the main objective(s) of the study, and state any hypotheses to be tested.	4
Methodology			
Study Design and Fundamentals	5	Provide precise details with respect to the design and fundamentals of the study, including but not limited to the following: Study design: prospective, retrospective, cross-sectional, etc. Study type: cadaveric (e.g. formalin fixed or fresh frozen), imaging, intraoperative, etc.	5
Setting	6	Describe clearly the location where the study was conducted and dates (month/year) between which the data were collected.	5
Sample Size	7	When appropriate, statistical power analysis should be used to calculate sample size or effect size. If relevant, justification for the study sample size should be briefly stated.	5
Subjects	8	Define clearly the eligibility criteria and methods of subject selection and inclusion, with details of the baseline and demographic selection criteria of the subjects (age, sex, healthy or diseased etc.) included in the study.	9
Reference Standard	9	Define clearly and accurately all anatomical definitions (normal anatomy, variations, classifications, etc.) by which data will be collected, analyzed, and compared. Citations should be included when appropriate.	6
Outcomes and/or Parameters	10	Define clearly the outcomes and parameters (e.g. prevalence of a variation, mean length and diameter of a structure, etc.) assessed in the study. When present, confounders should be clearly stated.	6-7
Measurement and Assessment	11	Indicate clearly the group of subjects included in each measurement/assessment (source of data). Provide clear details about the methods of measurement/assessment of each outcome and/or parameter (e.g. reference points for length measurements, internal or external diameter, etc.).	7
Modality	12	Describe clearly the materials, equipment, and instruments used (with manufacturer/supplier details) to conduct the specific study design.	5-7
Technique	13	Describe precisely the methods (e.g. dissection technique, image reconstruction, etc.) applied in the study to allow for reproducibility. Relevant details (profession, years of experience) regarding the individual(s) performing the technical aspect of the study are recommended.	5
Bias	14	Identify any potential source of bias and, when present, describe measures implemented to assess the risk of bias.	N/A
Statistical Approach	15	Describe all statistical methods for analyzing the data, including those of confounders. Statistical methods for additional analyses (e.g. subgroup/sensitivity analyses), when performed, should be described.	7-8
Ethics	16	Provide the details of compliance with ethical guidelines, including the name of the review board or agency, approval number, and date. AQUA endorses the Helsinki Declaration and its later amendments. When appropriate, details of written, informed consent should be clearly stated.	8

Results			
Subjects	17	Report the numbers of subjects included in the study, including data on their baseline and demographic characteristics. When needed, provide reason(s) and data on characteristics of the subjects excluded from the study at any stage.	9
Main Results	18	Provide unaltered/non-manipulated summary data (number [percentage]) or estimates (with confidence intervals and values of consistency when applicable) from the analyses performed. Tabular presentation of the results is highly recommended.	10-11
Descriptive Anatomy	19	Present clear and comprehensible figures (i.e. images, illustrations, diagrams, etc.), labeled as appropriate, to explain the results where needed AND describe clearly any anatomical findings that could be ambiguous, questionable, or atypical. New classifications of anatomical variations should be complemented by representative figures and corresponding dissection/imaging photographs.	Figures
Confounders	20	Present precise data from assessment/measurement of confounders, if any.	N/A
Additional analyses	21	Provide clear results of additional analyses (e.g. subgroup/sensitivity analyses), if performed. Tabular presentation of the results is highly recommended.	Tables
Discussion			
Key Findings	22	Include summary of key evidence/findings from the study pertaining to the rationale/objectives of the study. No new study results should be presented in the discussion.	12-13
Interpretation and Comparison(s)	23	Provide comprehensive (but judicious) interpretation of the results from the study, and comparison and/or reference to the results from other studies on the topic, appropriately cited. Meaningful clinical impact/significance of the findings from the study should be discussed where relevant.	12-16
Implication(s)	24	State briefly the implications of the findings or potential areas of the study that require further research.	16-17
Limitation(s)	25	Discuss briefly limitations of the study at any stage, including risk of bias, potential confounders, or intraobserver and/or interobserver variability.	17
Conclusions			
Summary	26	Summarize the key information (i.e. “take-home message”) directly supported by the findings/evidence from the study.	17
Other Information			
Acknowledgement	27	Acknowledge individual(s), institution(s), or third parties who significantly contributed to the study.	18
Conflict of interest	28	Disclose any conflicts of interests related to the study for all contributing authors.	18
Funding	29	Describe sources of funding for the study and any other support.	18

* If an item is not applicable to the study, mark N/A in the page number box.

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