INSTITUTE OF PREVENTIVE PEDIATRICS
FACULTY OF SPORTS AND HEALTH SCIENCES

TECHNICAL UNIVERSITY OF MUNICH

Cardiovascular Prevention in Childhood

Assessment of Carotid Intima-Media Thickness and Arterial Distensibility in Children and Adolescents (7 - 17 years) and the Relation of Vascular Status to Health-Related Fitness

Heidi Weberruß

Vollständiger Abdruck der von der Fakultät der Sport- und Gesundheitswissenschaften der Technischen Universität München zur Erlangung des akademischen Grades eines

Doktors der Philosophie (Dr. phil.)
genehmigte Dissertation

Vorsitzender: Prof. Dr. Jürgen Beckmann

Prüfer der Dissertation:

1. Prof. Dr. Renate Oberhoffer
2. Prof. Dr. Arno Schmidt-Trucksäss (Universität Basel, Schweiz)

Die Dissertation wurde am 23.02.2016 bei der Technischen Universität München eingereicht und durch die Fakultät der Sport- und Gesundheitswissenschaften am 07.11.2016 angenommen.

TABLE OF CONTENTS

I ABBREVIATIONS

AC	arterial compliance
β	stiffness index β
%BF	percentage of body fat
BMI	body mass index
BP	blood pressure
CA	carotid artery
CCA	common carotid artery
cIMT	carotid intima-media thickness
CVD	cardiovascular disease
IMT	intima-media thickness
Ep	elastic modulus
PACER	progressive cardiovascular endurance run
PP	pulse pressure
PWV	pulse wave velocity
PWV β	local pulse wave velocity β
SDS	standard deviation score
WC	waist circumference
WHR	waist-to-hip ratio
WHtR	waist-to-height ratio

II LIST OF FIGURES

III LIST OF TABLES

1 INTRODUCTION

1.1 GENERAL INTRODUCTION AND STUDY PURPOSE

Atherosclerosis is a slowly progressing disease caused by inflammatory processes within the arterial wall [1]. It is characterized by a silent subclinical phase that onsets in early childhood and represents the underlying mechanism for cardiovascular disease (CVD) [2, 3], the leading cause of morbidity and mortality in western countries [4].

Structural alterations of the arterial wall are characterized by an increased carotid intima-media thickness (cIMT), and functional alterations by impaired arterial distensibility. Both of them are early signs in the atherosclerotic process and reliable subclinical markers of the disease [5-7]. cIMT and arterial distensibility can easily and non-invasively be assessed by B- and M-mode ultrasound.

Already in childhood there are pathological conditions, which lead to a more pronounced increase in cIMT and to an already increased cardiovascular risk at a young age. These conditions are familial hypercholesterolemia [8-10], hypertension [11, 12], obesity [13, 14], metabolic syndrome [15], and type I diabetes [16, 17]. Even earlier as cIMT increases, distensibility can be impaired under conditions such as familial hypercholesterolemia [18], hypertension [19], type I diabetes [20], and obesity [21].

Reference values for cIMT and arterial distensibility in children and adolescents exist [22-25], but are difficult to apply. Values depend strongly on the respective measurement protocol and device used for measurement [26]. The aim of this study was therefore to assess cIMT and arterial distensibility in a large pediatric cohort (n = 1017) aged 7-17 years, to establish reference values with a new ultrasound device (ProSound Alpha 6; Aloka/ Hitachi Medical Systems GmbH, Wiesbaden, Germany), that measures cIMT and arterial distensibility in one examination at the same vessel segment.

Low physical fitness is an independent risk factor for future CVD [27-29]. For this reason, children's health-related fitness was additionally assessed to examine if low physical fitness is already associated with an increased cIMT and impaired arterial distensibility at a young age.

1.2 Subclinical Atherosclerosis in Children and Adolescents

Atherosclerosis is a chronic, inflammatory disease of the arterial intima layer. It is a dynamic process, affecting the entire arterial tree [30, 31]. Atherosclerosis is the most common arterial disease [32] and leading cause of morbidity and mortality amongst men and women of 60 years and older – but it is not a complete inevitable consequence of ageing [31]. The disease starts in early childhood [1, 31, 33-35] or even as early as the fetal stage with formation of intimal cell masses [36]. Not all elderly individuals develop atherosclerosis. Under certain conditions, however, the disease may occur in adolescents [37] or develop in early adulthood, promoted by risk factors during childhood [38]. Atherosclerotic end points are myocardial infarction, stroke or ischemic diseases like angina pectoris or peripheral artery disease (Figure 1).

Up to date, the initial step leading to atherosclerosis is not fully understood. There are two hypotheses explaining atherogenesis: First, the Response-to-Injury Hypothesis by Ross [39] and second, the Lipoprotein-induced Hypothesis by Goldstein [40]. Ross refers to von Rokitansky's description of intimal thickening by fibrin deposition and lipid accumulation [41] and Virchow's view on inflammatory processes being responsible for intimal thickening [42]. Goldstein focused on the interaction of oxidized LDL cholesterol and macrophages initiating the disease [40]. In current research, endothelial dysfunction is known to play a further important role in atherogenesis [43].

Fatty streaks are regarded as initial atherosclerotic lesion [44], described as flat intimal thickening, without structural remodeling and restriction of the respective artery. Fatty streaks are not unlikely to be present in the aorta of children between 1-15 years [1, 33, 34], and occur about 5-10 years later in the coronary arteries [45].

Fibrous plaques are elevated intimal lesions, containing different amounts of extracellular lipids and cholesterol esters with a collagenous or muscular cap, affecting the arterial structure [45]. In autopsy studies, fibrous plaques were first detected in the second decade of life, and increases in frequency and extent throughout the third and fourth decade [2]. According to McGill [45], the process from fatty streaks into fibrous plaques is a continuous development with varying lesion stages in between harmless fatty streaks and fibrous plaques. Transformation of fatty streaks into fibrous plaques is promoted and accelerated by presence of risk factors [35, 46].

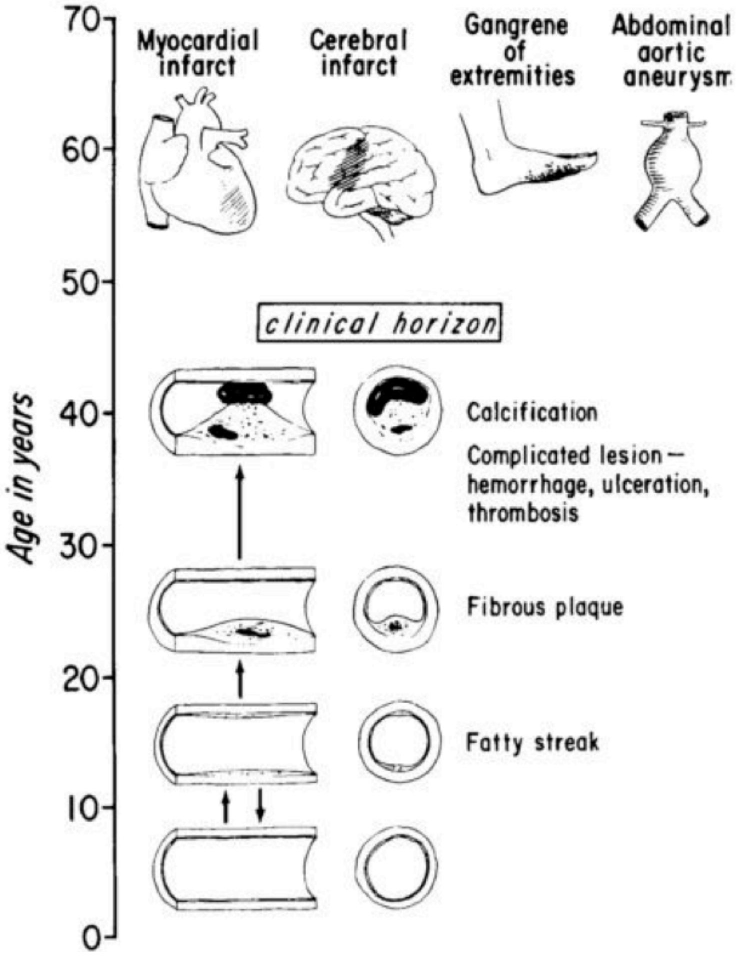

Figure 1. Atherogenesis from early development of fatty streaks in childhood to cardiocerebral endpoints in adults, by Strong, J.P. The natural history of atherosclerosis in childhood. Ann NY Acad Sci (1991). 623: 9-15.

Since the 1950s, several study groups were engaged in identification of risk factors linked to atherosclerosis and hence a higher risk for future cardiovascular disease in adults and children [34, 46-48]. These risk factors (Table 1) can be divided into traditional non-modifiable [34, 46-48] or traditional modifiable risk factors [49-55], and emerging or new risk factors, respectively [56, 57]. The latter still need to be further investigated [57].

In the advanced atherosclerotic stage, fibrous plaques restrict the arterial lumen, leading to a diminished perfusion or an occlusion of the artery with subsequent ischemia of the target organ. Before this clinical manifestation of atherosclerosis, subclinical markers like intima-media thickness (IMT) of arterial walls and arterial distensibility can be assessed [2, 58].

Table 1. Traditional and emerging risk factors for cardiovascular disease.

Traditional Risk Factors		**Emerging Risk Factors**
non-modifiable	modifiable	
family history	dyslipidemia	metabolic syndrome
male sex	type II diabetes mellitus	high sensitive CRP
age	smoking	fibrinogen
	sedentary lifestyle	homocysteine
	overweight/ obesity	lipoprotein a
		infections
		psychosocial factors

1.3 Carotid Intima-Media Thickness and Arterial Stiffness as Parameters to identify Subclinical Atherosclerosis

The intima and media wall complex of an artery, combined as IMT represents a reliable, surrogate marker for early subclinical atherosclerotic changes of the arterial wall [59, 60]. In 1986, Pignoli et al. [61] demonstrated a strong correlation between the common carotid artery (CCA) far wall IMT, assessed via ultrasound, and histological findings. The association between an increased cIMT and atherosclerotic risk factors has been described in several studies [62].

cIMT assessment via B-Mode ultrasound is a safe, inexpensive and quick method to non-invasively assess IMT in different subclinical stages of atherogenesis, and to monitor progression of the disease or results of pharmacological or risk factor modifying interventions. Located superficially on the neck, the carotid artery is further easy to access and represents a feasible method [63]. Figure 2 shows an ultrasound image with far wall IMT depicted as two echogenic lines, representing the lumen-intima and media-adventitia interface. Near wall IMT, however, is not a true measure of IMT but a composition of the adventitia and media layer with parts of the intima [64].

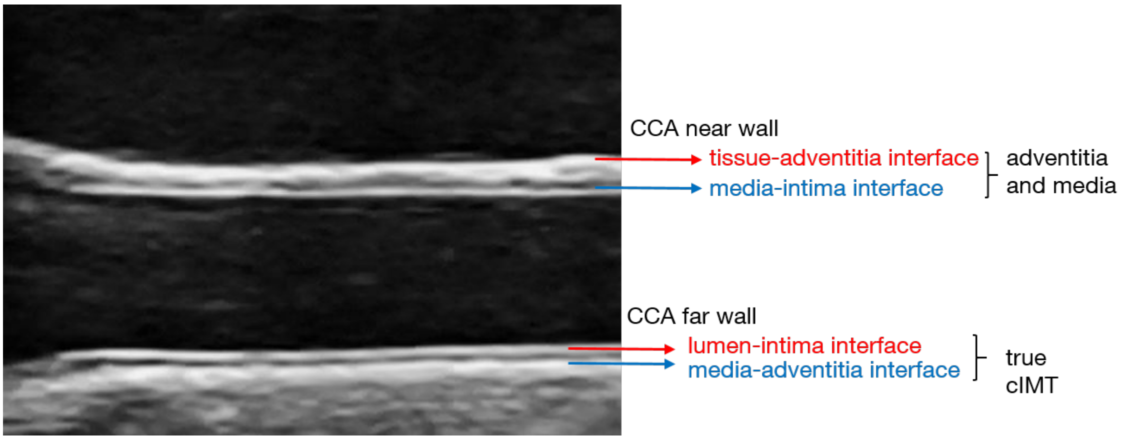

Figure 2. Ultrasound image of the common carotid artery (CCA) with depiction of true carotid intima-media thickness (cIMT), detectable at the vessel far wall. Ultrasound reflection at the near wall does not represent true cIMT but thickness of the adventitia and media with parts of the intimal layer.

In adults, IMT differs with sex and race [65, 66], and increases physiologically with age [65, 67, 68]. In children, an age-dependent increase of cIMT is discussed controversially [24, 25, 67]. Values above the sex- and age-adjusted 75th percentile are considered abnormal in adults. The 75th percentile as cut-off value is also applied in children [66, 69].

cIMT reference values assessed in children vary much depending on the measuring protocol and ultrasound device used [26]. Table 2 displays existing reference values for children and adolescents, including results of this study [70]. With the device used in this study (ProSound Alpha 6; Aloka/ Hitachi Medical Systems GmbH, Wiesbaden, Germany), reference values for children and adolescents did not yet exist.

Comparison of cIMT in patient groups compared to healthy controls revealed significant higher values in children with hypercholesterolemia [71, 72], hypertension [11, 12, 19], type I diabetes [16, 73], obesity [13, 14], and in young adults with the metabolic syndrome [74].

Table 2. Reference values for carotid intima-media thickness (cIMT) in children and adolescents

Study	Subjects	Age (years)	cIMT
Böhm et al. [25]	n = 267	6-17	0.49 - 0.55 mm
Doyon et al. [23]	n = 1155	6-18	0.36 - 0.4 mm
Jourdan et al. [22]	n = 247	10-20	0.38 - 0.4 mm
Sass et al. [24]	n = 160	10-18	0.48 - 0.5 mm
Weberruss et al. [70]	n = 673	6-17	0.38 - 0.54 mm

Even earlier than the artery's structure, functional properties are affected, characterized by impaired arterial distensibility [3, 75]. In this study, distensibility is used as umbrella term for arterial compliance (AC) and its reciprocal, arterial stiffness. Distensibility refers to the artery's ability to cushion the arterial pressure wave during its propagation through the arterial system. It is determined by visco-elastic properties of the arterial wall [76, 77].

Arterial distensibility can be defined with several parameters (elastic modulus, Young’s modulus, arterial distensibility, arterial compliance, pulse wave velocity, augmentation index, stiffness index β, capacitative compliance, and oscillatory compliance) that refer to different aspects of the artery [7], which will not be further discussed in this work. Distensibility parameters in this study were AC, elastic modulus (Ep), stiffness index β (β), and local pulse wave velocity β (PWV β).

Arterial distensibility can also be assessed non-invasively by M-mode ultrasound wall tracking techniques, measuring changes in lumen diameter in response to blood pressure changes during the cardiac cycle [59, 78]. Comparable to cIMT, distensibility parameters strongly differ between the protocol and device used for assessment. For the device applied in this study, comparable references in children do not exist, except for unpublished values by the device producer, assessed in the Chinese population (Aloka/ Hitachi 2008). References by Jourdan et al. [22] or Doyon et al. [23] can be considered for β (Table 3).

Increased arterial stiffness is already present in children with heterozygous familial hypercholesterolemia, hypertension, the metabolic syndrome, type I diabetes or severe obesity [15, 18, 19, 79]. Higher arterial stiffness leads to an increased left ventricular afterload resulting in left ventricular hypertrophy and diminished coronary perfusion [80].

Table 3. Reference values for arterial distensibility parameters in children and adolescents.

Study	Subjects	Age (years)	AC	Ep	β	PWV β (m/s)
Doyon et al. [23]	n=1155	6-18			1.6-4.7	
Jourdan et al. [22]	n=247	10-20			3.25-3.5	
Aloka/ Hitachi [81]	n=282	<10-19	2.1-1.43	24.97-44.42	2.7-4.13	3.1-4.29
Weberruss et al. [70]	n=870	6-17	0.59-1.9	25.7-72.7	2.2-5.9	3.1-5.2

AC = arterial compliance; Ep = elastic modulus; β = stiffness index β; PWV β = local pulse wave velocity β.

1.4 Health-Related Fitness in Association to Atherosclerosis and Cardiovascular Disease

Physical fitness describes a large set of attributes, being differently associated with atherosclerosis or cardiovascular disease. In this study physical fitness was assessed as health-related fitness (HRF), using the test battery FITNESSGRAM® (The Cooper Institute, Dallas, TX, United States). FITNESSGRAM® was chosen because it is a simple and inexpensive test battery, consisting of six tasks, which do not require computer based or other technical equipment. The tasks are easily understood by pupils of all ages and require a mean testing time of only about 15 minutes [82]. HRF focuses on the components aerobic or cardiopulmonary capacity, respectively, on strength, and flexibility [83]. The term fitness is generally used to express aerobic or cardiopulmonary capacity and will be applied in this study, too.

Fitness is strongly and independently associated with cardiovascular health [27-29]. Fit adults with a high aerobic capacity have a reduced risk for cardiovascular disease independent of other risk factors. Poor aerobic capacity on the other hand is one major cardiovascular risk factor [84]. Atherosclerosis prevalence among fit persons is lower compared to unfit subjects. Progression of atherosclerosis can be retarded by increasing someone’s fitness level [85, 86]. These benefits can already be seen at a young age, as fit children have a lower cardiovascular risk [87] and lower risk for the metabolic syndrome than unfit controls [88]. Benefits of high fitness levels in childhood remain into adulthood [89].

Regarding aerobic capacity and cIMT, controversial results exist, with most studies reporting no significant association between aerobic capacity and cIMT in children [90, 91] and within the adult population [92-95]. Regarding arterial stiffness, there is a significant inverse association between aerobic capacity and arterial stiffness and a positive association with compliance in children and adults [91, 95-98].

Fewer studies investigated the influence of muscular strength on cardio-vascular risk factors and arterial parameters. Melo et al. [99] reported a significant inverse relationship of strength and cIMT in children, others a positive influence on traditional cardiovascular risk factors [89, 100-102]. In adults, resistance training positively modifies traditional cardiovascular risk factors [103], but no significant association could be found between strength and cIMT [104].

Flexibility was assessed as part of FITNESSGRAM® but not taken into account for further analysis. In the current literature, flexibility does not show any significant associations with cardiovascular health [89, 105]. This study therefore focuses on aerobic capacity as primary outcome, and secondary on parameters of muscular strength.

2 METHODOLOGY

2.1 PREVENTION PROJECT “STERNSTUNDEN DER GESUNDHEIT”

The prevention project “Sternstunden der Gesundheit“ took place between October 2012 and July 2013 as cooperation between the Technical University of Munich, Institute of Preventive Pediatrics, and the Ludwig-Maximilians-Universität München, Department of Pediatric Cardiology, the non-profit organization “Aktion Sternstunden e.V.” and the district office “Berchtesgadener Land”. All schools of the district “Berchtesgadener Land” in southern Bavaria were invited to participate. Participation was voluntary and only allowed after written informed consent form was signed by parents and parents and children $\geq$ 14 years.

All examinations took place in the same facility, following a standardized protocol, at the same time on two days a week. The test team consisted of 10 persons, two trained ultrasound examiners, and eight sports students who were responsible for anthropometric measurements and HRF testing. The test team examined one class of about 20 - 30 pupils a day, which were divided into small subgroups with 4 - 6 pupils each. Within these sub-groups, pupils passed the ultrasound examination (cIMT, arterial distensibility, and BP) followed by HRF testing.

2.2 STUDY PARTICIPANTS

A total of 1017 children (483 boys/ 534 girls), 11.9 ± 2.35 years of age, participated in the prevention project. Anthropometric data of study participants are displayed in Table 4 and 5 separately for boys and girls and split into five age groups.

Table 4. Anthropometric characteristics for male study participants in different age groups.

AGE GROUP [years]	6 - 7.99	8 - 9.99	10 - 11.99	12 - 13.99	14 - 15.99	16 - 17.99
SUBJECTS	n = 5	n = 113	n = 191	n = 104	n = 50	n = 20
AGE [years]	7.8 ± 0.13	9.0 ± 0.54	11.1 ± 0.53	12.7 ± 0.55	14.8 ± 0.56	16.6 ± 0.46
HEIGHT [cm]	132.2 ± 3.99	135.7 ± 6.35	148.1 ± 7.26	155.9 ± 8.90	171.8 ± 8.68	176.4 ± 6.25
WEIGHT [kg]	30.4 ± 2.03	32.1 ± 6.88	40.9 ± 8.62	47.6 ± 12.51	62.5 ± 14.76	67.5 ± 14.03
BMI [kg/m^2]	17.2 ± 1.26	17.3 ± 2.71	18.5 ± 3.03	19.3 ± 3.63	21.0 ± 3.98	44.6 ± 3.39
BMI SDS	0.1 ± 0.83	0.2 ± 0.99	0.2 ± 0.99	0.0 ± 1.15	0.2 ± 14.76	21.6 ± 11.18

BMI = Body Mass Index; SDS = Standard Deviation Score

Table 5. Anthropometric characteristics for female study participants in different age groups.

AGE GROUP [years]	6 - 7.99	8 - 9.99	10 - 11.99	12 - 13.99	14 - 15.99	16 - 17.99
SUBJECTS	n = 7	n = 108	n = 146	n = 108	n = 129	n = 36
AGE [years]	7.6 ± 0.35	9.0 ± 0.53	11.0 ± 0.54	12.8 ± 0.61	14.9 ± 0.59	16.7 ± 0.52
HEIGHT [cm]	129.8 ± 4.64	135.0 ± 7.49	148.3 ± 7.96	158.2 ± 7.63	165.0 ± 5.74	165.1 ± 6.09
WEIGHT [kg]	29.8 ± 6.74	31.4 ± 6.98	40.2 ± 10.36	47.4 ± 9.07	57.1 ± 12.11	61.2 ± 11.56
BMI [kg/m^2]	17.6 ± 3.27	17.1 ± 2.65	18.1 ± 3.53	18.8 ± 2.78	20.9 ± 4.06	41.9 ± 4.37
BMI SDS	0.6 ± 1.20	0.1 ± 1.03	0.0 ± 1.13	-0.2 ± 0.96	0.1 ± 12.11	22.5 ± 11.61

BMI = Body Mass Index; SDS = Standard Deviation Score

Children of 13 schools, 10 different grades and 6 different types of schools participated in the project (Figure 3 and 4). Children with any kind of chronic disease, acute infection or orthopedic complaints were excluded from study participation.

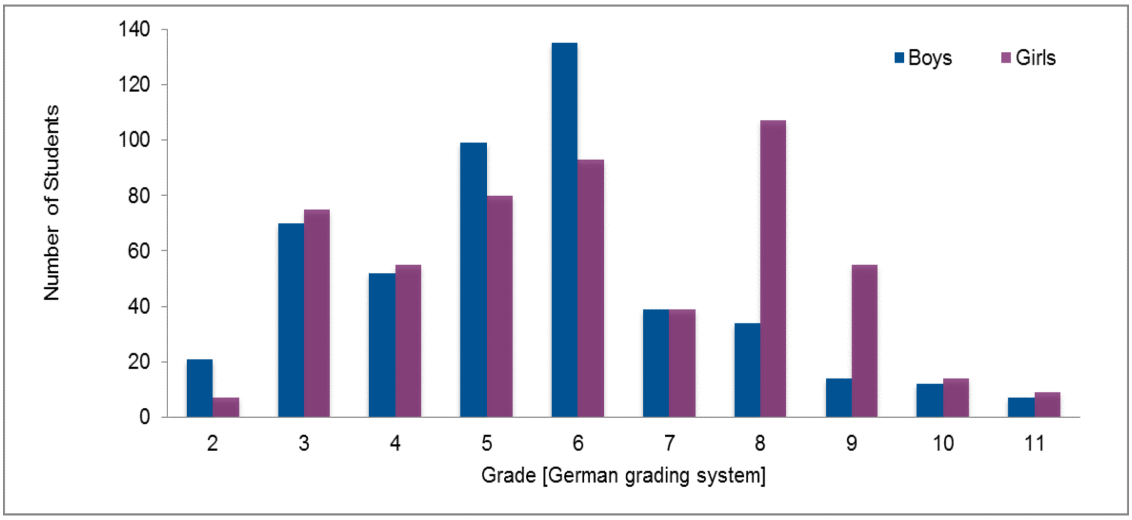

Figure 3. Distribution of participating boys and girls per grade.

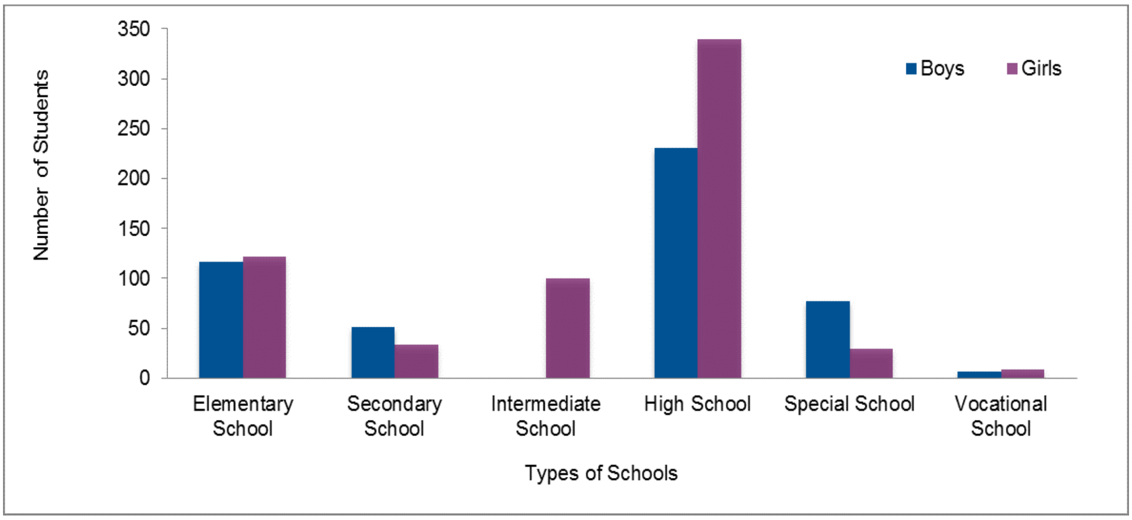

Figure 4. Distribution of participating boys and girls per type of school. Elementary school = Grundschule, Secondary School = Mittelschule, Intermediate School = Realschule, High School = Gymnasium, Special School = Förderschule, Vocational School = Berufsfachschule.

Due to technical problems with the ultrasound storage function, IMT data of 281 and arterial distensibility data of 89 subjects were lost. Eight children were excluded from participation in HRF testing (injuries or no proper sportswear). As consequence, study results were obtained in four different sub-samples and will further be discussed separately for cIMT (n = 690), arterial distensibility (n = 870), and cIMT and arterial distensibility (n = 656). Ultrasound and HRF data were investigated in n = 697 children (376 girls). All sub-samples are displayed in Figure 5.

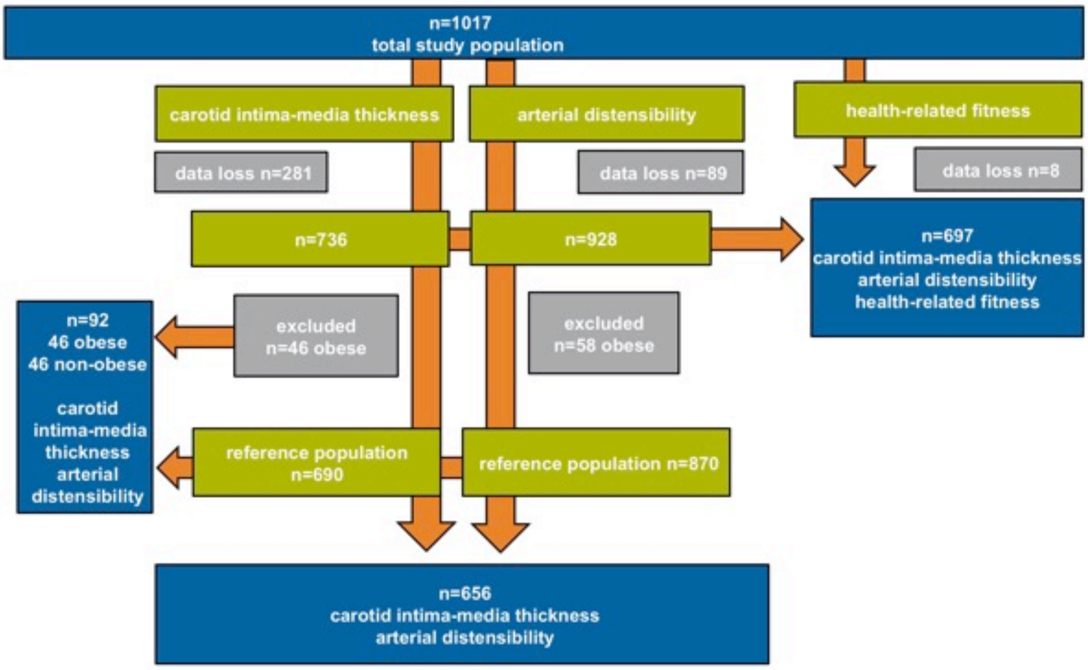

Figure 5. Overview of the entire study population (n = 1017) and respective sub-samples.

2.3 Anthropometry and Blood Pressure

Body weight and height, waist and hip circumference were measured according to standardized guidelines [106] to the nearest 0.1 kg or 0.1 cm, respectively (seca 799; seca, Hamburg, Germany). Body mass index (BMI) was calculated as body weight (kg)/ height^2 (m^2). Standard deviation scores (SDS) for BMI were calculated and weight categories defined according to German reference values [107]. Blood pressure (BP) was measured oscillometrically on the left arm (Mobil-O-Graph ®; I.E.M., Stolberg, Germany) after participants rested for 10 minutes in supine position. As only one BP measurement was performed, children were not defined with manifest hypertension but with suspected hypertension (BP > 95th percentile). SDS for BP were calculated according to German reference [108].

2.4 Carotid Intima-Media Thickness and Arterial Distensibility

cIMT and arterial distensibility were assessed by semi-automated B- and M-Mode ultrasound, respectively, with a high frequency linear array probe of 5-13 MHz (ProSound Alpha 6; Aloka/ Hitachi Medical Systems GmbH, Wiesbaden, Germany) by two trained examiners. The semi-automated measurement technique combines automated edge detection with manual correction. cIMT was measured in B-Mode according to the Mannheim Consensus [109] on CCA far wall on the left and the right side (Figure 6). Children were examined in supine position, after 15 minutes of rest, the neck slightly extended and their head turned 45° opposite the site being scanned. On each side, two measurements were performed, 1 cm proximal to the bulb at end-diastolic moment (R-wave), when cIMT is thickest [110]. The cardiac cycle was controlled with a three-lead ECG. cIMT was calculated as average mean value out of four measurements.

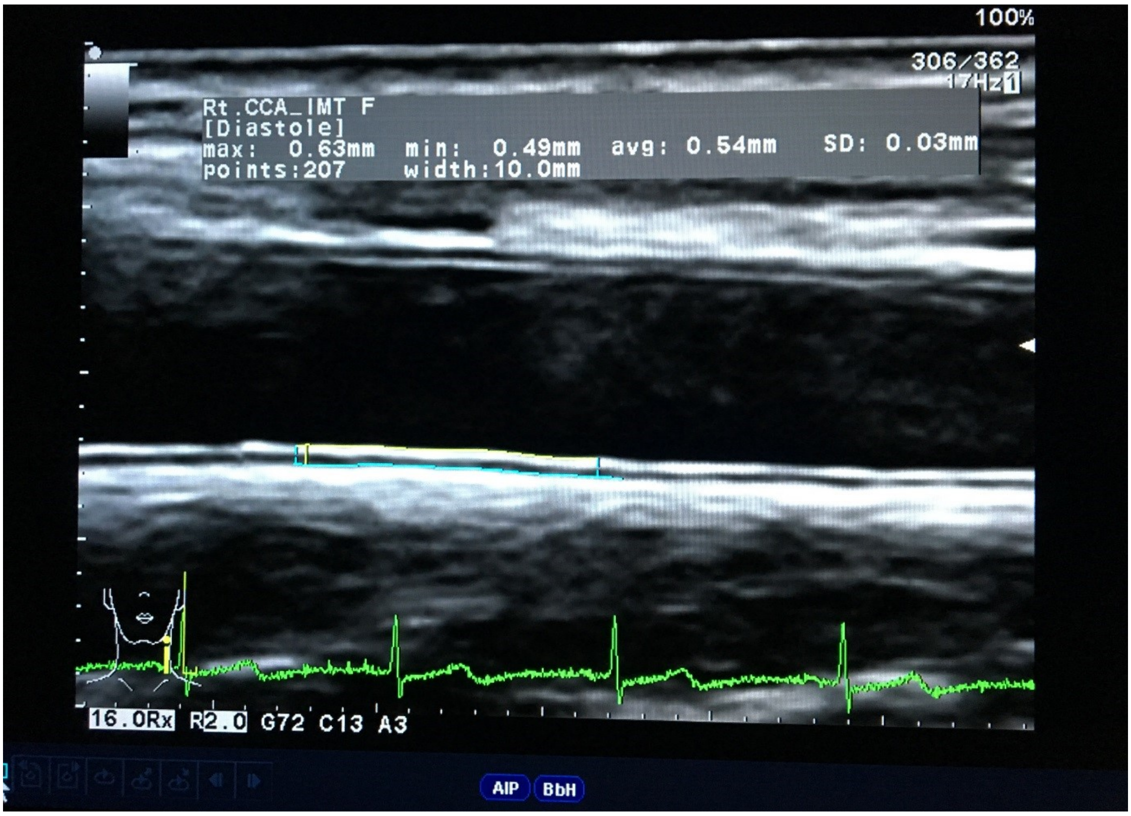

Figure 6. Ultrasound image of a carotid intima-media thickness (cIMT) measurement at the left common carotid artery far wall, 1 cm proximal to the bulb at end-diastolic moment (R-Wave).

In this study, arterial distensibility was assessed as arterial compliance (AC), and following stiffness indices: elastic modulus (Ep), stiffness index β (β), and local pulse wave velocity β (PWV β). Arterial compliance (AC) indicates the artery's ability to respond to changes in blood volume [111]. Ep, β, and PWV β increase with increasing arterial stiffness but differ according to blood pressure dependency, with being strongly dependent on BP (Ep) or less (β, PWV β) [22].

Distensibility parameters were assessed in real-time M-Mode via radio frequency echo-tracking over 5 consecutive cardiac cycles at the same location that cIMT has been measured. Therefore, two tracking gates were placed on the CCA near and far wall IMT-complex, to automatically follow vessel motion and calculate changes in diameter from systole to diastole by radio frequency signal, providing an accuracy of 0.01mm resolution at 10 MHz transmission-reception rate (Figure 7).

As arterial distensibility depends on blood pressure, one BP measurement was taken on the left arm and applied into the calculation. For each side, two video loops were stored and parameters calculated according to the formulae below (1-4), where D is the change in blood vessel cross-sectional area.

$$(1)\ AC = \pi(D_{max^2} - D_{min^2})/[4(BP_{max} - BP_{min})]$$

$$(2)\ Ep = (BP_{max} - BP_{min})/[(D_{max} - D_{min})/D_{min}]$$

$$(3)\ \beta = ln(BP_{max} / BP_{min})/[(D_{max} - D_{min})/D_{min}]$$

$$(4)\ PWV\beta = \sqrt{}((\beta * BP_{min})/(2\rho))$$

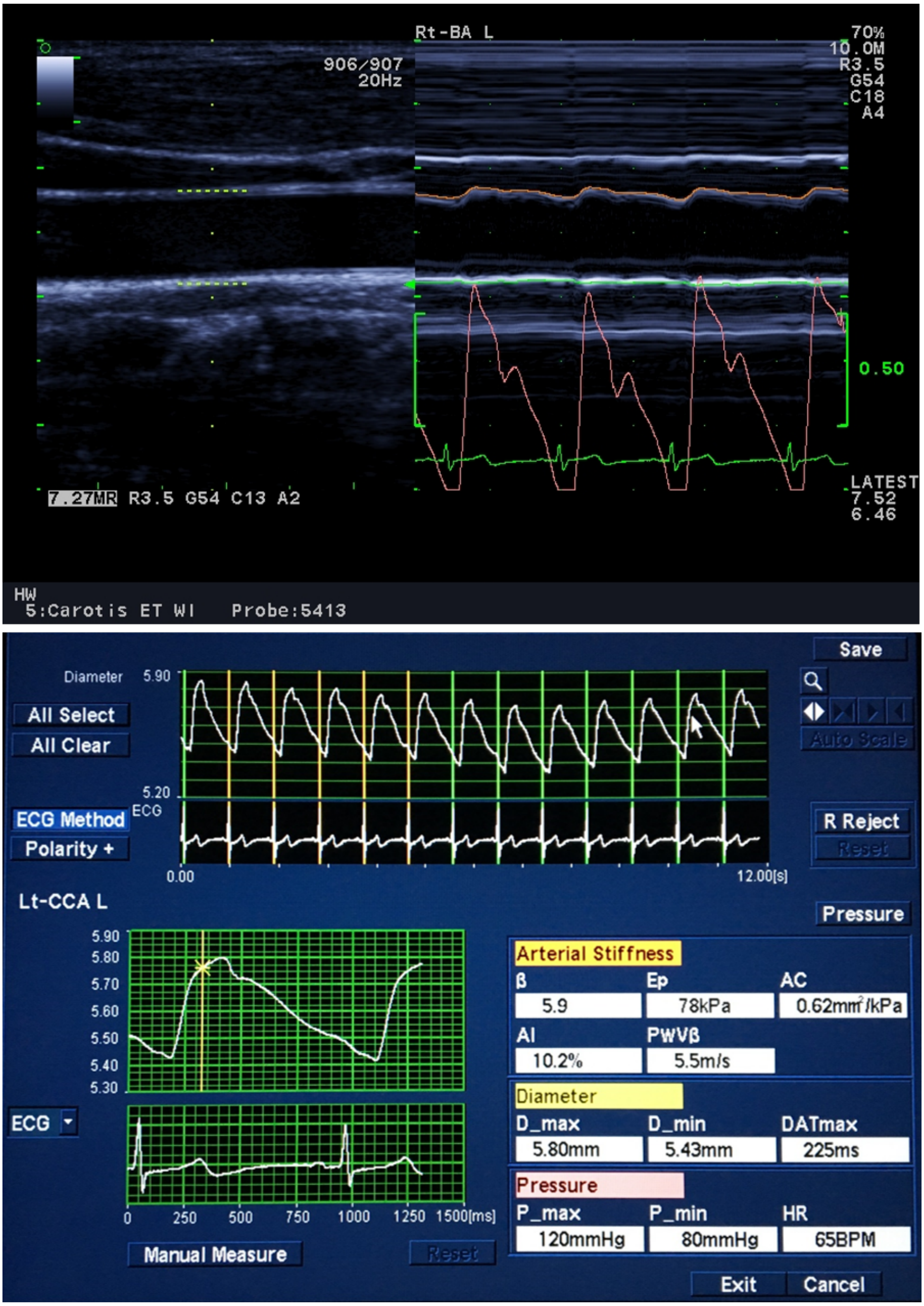

Figure 7. Echo tracking measurement of common carotid artery distensibility (above) and analysis of arterial compliance (AC), elastic modulus (Ep) stiffness index β (β), and local pulse wave velocity β (PWV β) (below).

2.5 Health-Related Fitness Testing

Health-related fitness was assessed using the test battery FITNESSGRAM® (The Cooper Institute, Dallas, TX, United States). The different motor tasks were (1) push-ups, (2) curl-ups, (3) trunk lift, (4) back saver sit and reach, (5) shoulder stretch, and (6) PACER (Figure 8). HRF testing was performed one by one following standardized instructions [82]. For further analysis with vascular data, only push-ups, curl-ups, and PACER were considered.

Figure 8. Health-related fitness testing with the test battery FITNESSGRAM®: (1) push-ups, (2) curl-ups, (3) trunk lift, (4) back saver sit and reach, (5) shoulder stretch, and (6) PACER.

(1) Push-ups measure strength and endurance of the upper body. Out of the starting position, a push-up with extended arms and legs, hands positioned vertically under the shoulders, the student lowers his body until a 90° elbow flex, and performs a push-up back into the starting position. The aim is to perform as many push-ups as possible. The test is terminated with the second incorrect push-up.

(2) Curl-ups measure strength and endurance of the abdominal muscles. The student lies in supine position on a gymnastic mat, his head touching the mat, legs bent, feet flat on the mat, and arms extended with fingertips

pointing to the toes. The student lifts his upper body, until his arms reach a mark on the gymnastic mat and returns back to the starting position. The aim is to perform as many curl-ups as possible. The test is terminated with the second incorrect curl-up.

(3) Trunk lift measures the static strength of lower back muscles. Starting position is prone on the mat, arms extended and hands placed under the thighs. The student lifts his upper body as high as possible and holds this position until the height is being measured as vertical distance between chin and floor in cm. The best attempt out of two is being recorded.

(4) The back saver sit and reach measures hamstring flexibility for the left and right leg, respectively. The student sits on a mat, one leg extended, the other one is bent. With the back kept straight, the student tries to reach with both hands for his toes. The distance between fingertips and toes is being recorded in cm.

(5) The shoulder stretch tests the upper arm and shoulder girdle flexibility for the left and right side, respectively. The student tries to touch his hands behind the back by reaching over the shoulder with one hand and under the shoulder with the other hand. The distance between the metacarpal bones was being recorded in cm.

(6) PACER is the acronym for progressive aerobic cardiovascular endurance run. It is a 20 meter shuttle-run at increasing speed. Within the first level, the time interval for a 20 meter lap is 9 seconds. Every minute, time to pass one 20 meter lap is reduced by 0.5 seconds, which refers to a starting speed of 8 km/h and an increase of about 0.5 km/h per minute. The speed is controlled by audio signals that terminate each lap. The student tries to run as many 20 meter laps as possible. The test is terminated by the second failure to reach the end of a 20 meter lap within the time limit.

3 PUBLICATIONS

3.1 INCREASED INTIMA-MEDIA THICKNESS IS NOT ASSOCIATED WITH STIFFER ARTERIES IN CHILDREN

HEIDI WEBERRUß, RAPHAEL PIRZER, BIRGIT BÖHM, JULIA ELMENHORST, ROBERT DALLA POZZA, HEINRICH NETZ, RENATE OBERHOFFER

ATHEROSCLEROSIS 242 (2015) 48-55

Subclinical atherosclerosis can be assessed via sonographic measurement of intima-media thickness and carotid artery distensibility, both may already be pathologically altered in childhood. Therefore, the purpose of this study was to provide reference percentiles and investigate possible associations between alterations of intima-media thickness and distensibility. Carotid intima-media thickness and distensibility was measured via B- and M-mode ultrasound. Distensibility was defined by arterial compliance, elastic modulus, stiffness index β, and local pulse wave velocity β. Age- and height-dependent reference values were calculated separately for boys and girls among 690 (intima-media thickness) and 870 (distensibility) non-obese children aged 7–17 years. Intima-media thickness and distensibility did not increase significantly with age or differ between boys and girls. Systolic blood pressure and body mass index were independent predictors of intima-media thickness, while an increased systolic blood pressure or pulse pressure was associated with stiffer arteries. Increased intima-media thickness was accompanied by higher arterial compliance and lower stiffness. Using this healthy cohort, we describe a functional and non-pathological arterial adaptation wherein an increase in intima-media thickness is not associated with stiffer arteries.

3.2 Intima-Media Thickness and Arterial Function in obese and non-obese Children

Heidi Weberruß, Raphael Pirzer, Birgit Böhm, Julia Elmenhorst,

Robert Dalla Pozza, Heinrich Netz, Renate Oberhoffer

BMC Obesity 2016 3:2

Obesity is an independent cardiovascular risk factor that contributes to the development of atherosclerosis. Subclinical forms of the disease can be assessed via sonographic measurement of carotid intima-media thickness (cIMT) and distensibility – both may already be altered in childhood. As childhood obesity increases to an alarming extent this study compares vascular data of obese with normal weight boys and girls to investigate the influence of obesity on cIMT and distensibility of the carotid arteries. cIMT and distensibility of 46 obese children (27 girls) aged 7-17 years were compared with measures of 46 sex- and age-matched normal weight controls. cIMT and distensibility were measured by B- and M-mode ultrasound and expressed as standard deviation scores (SDS). Arterial distensibility was defined by arterial compliance (AC), elastic modulus (Ep), stiffness index β (β) and local pulse wave velocity β (PWV β). Obese girls had significantly stiffer arteries compared with normal weight girls (Ep SDS 0±1.06 vs. 0.64±1.24, β SDS -0.01±1.06 vs. 0.6±1.17 $p<.01$, PWV β -0.12±1.05 vs. 0.54±1.2 $p<.05$). No significant differences were observed for boys. In multiregression analysis, BMI significantly influenced Ep, β and PWV β but not cIMT and AC. Obese girls seemed to be at higher cardiovascular risk than boys, expressed by stiffer arteries in obese girls compared with normal weight girls. Overall, BMI negatively influenced parameters of arterial stiffness (Ep, β, and PWV β) but not compliance or cIMT.

3.3 Reduced Arterial Stiffness in very fit Boys and Girls

Heidi Weberruß, Raphael Pirzer, Thorsten Schulz, Birgit Böhm,

Robert Dalla Pozza, Heinrich Netz, Renate Oberhoffer

Cardiology in the Young. 2016 Mar 29:1-8

Low cardiorespiratory fitness is associated with higher cardiovascular risk, whereas high levels of cardiorespiratory fitness protect the cardiovascular system. Carotid intima-media thickness and arterial distensibility are well-established parameters to identify subclinical cardiovascular disease. Therefore, this study investigated the influence of cardiorespiratory fitness and muscular strength on carotid intima-media thickness and arterial distensibility in 697 children and adolescents (376 girls), aged 7-17 years. Cardiorespiratory fitness and strength were measured with the test battery FITNESSGRAM®; carotid intima-media thickness, arterial compliance, elastic modulus, stiffness index β, and local pulse wave velocity β were assessed by B- and M-mode ultrasound at the common carotid artery. In bivariate correlation, cardiorespiratory fitness was significantly associated with all cardiovascular parameters and was an independent predictor in multivariate regression analysis. No significant associations were obtained for muscular strength. In a one-way variance analysis, very fit boys and girls (58 boys and 74 girls > 80th percentile for cardiorespiratory fitness) had significantly decreased stiffness parameters (expressed in standard deviation scores) compared with low fit subjects (71 boys and 77 girls < 20th percentile for cardiorespiratory fitness): elastic modulus -0.16±1.02 vs. 0.19±1.17, P=0.009; stiffness index β -0.15±1.08 vs.0.16±1.1, P=0.03; local pulse wave velocity β -0.19±1.02 vs.0.19±1.14 P=0.005. Cardiorespiratory fitness is associated with healthier arteries in children and adolescents. Comparison of very fit with unfit subjects revealed better distensibility parameters in very fit boys and girls.

4 Discussion

Main findings of this study were: (1) Age had no significant influence on cIMT ($p = .09$). (2) There were no significant differences in cIMT between boys and girls in the overall study population and for most age-groups. (3) Overweight and hypertensive children had significantly increased cIMT values compared to normal weight and normotensive subjects, whereas no significant differences occurred between the obese sub-sample ($n = 46$) and sex- and age-matched normal weight controls. (4) A higher BMI was associated with more elastic arteries, except for stiffer arteries in obese girls compared with normal weight controls. (5) cIMT was positively correlated with AC and inversely with Ep, β, and PWV β. Complemented by higher cIMT and AC, and lower Ep, β, and PWV β in fit children, we state a functional adaptation of the arterial wall and no pathological alteration.

4.1 Carotid Intima-Media Thickness: Heterogeneity in Measurement Protocols, and Influence of Sex, Age, and Body Weight

In the early 1990s, cIMT came up as popular tool, to evaluate early subclinical atherosclerosis [3, 65] and has become the most widely used non-invasive method to examine arterial wall changes (Figure 9) [5]. In children, where manifest atherosclerosis is unlikely to occur, cIMT has gained importance to detect early subclinical wall changes, as a feasible, non-invasive and non-painful examination method that is free from radiation exposure [63, 112].

This study provides cIMT reference values for boys and girls aged 7-17 years, assessed in the to-date largest German study cohort ($n = 690$) [70]. cIMT reference values, assessed by B-Mode ultrasound, have been published by different study groups [22-25, 67] but should be interpreted cautiously. There is large variation in cIMT values, due to differences in measurement protocols [113, 114].

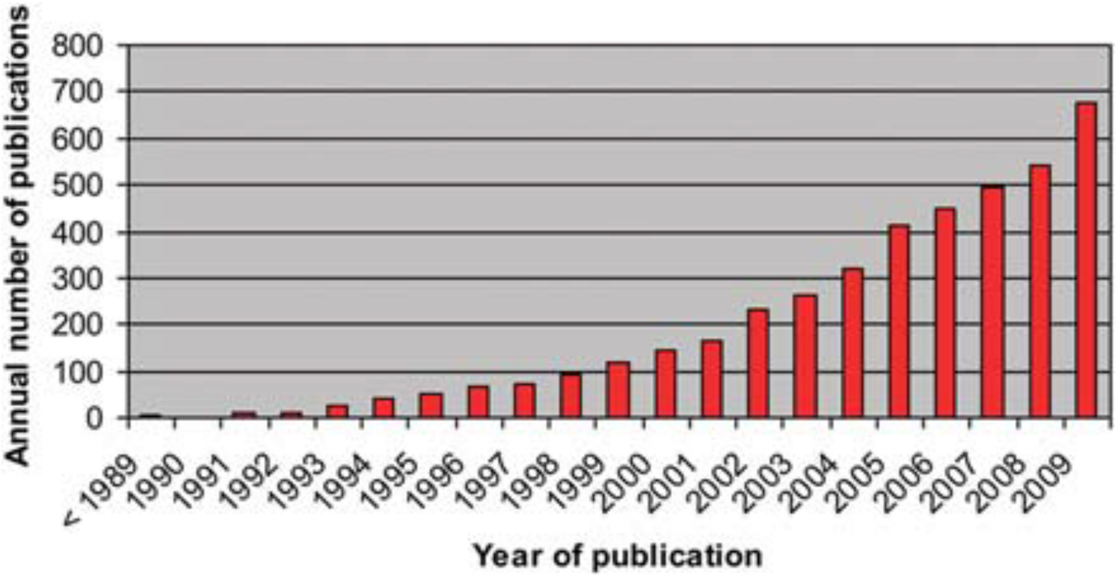

Figure 9. Rise in number of publications about carotid intima-media thickness, by O'Leary, D.H. and M.L. Bots, Imaging of atherosclerosis: carotid intima-media thickness. Eur Heart J, 2010. 31(14): p. 1682-9.

cIMT can be assessed at the CCA, internal CA or bifurcation, and, if measurement is performed at the CCA, this location is most differently defined in respect to its distance to the bulb [26]. Further differences in measurement protocols include the wall side obtained (near or far wall or both), and measuring technique applied (manual caliper method, automated edge-detection or semi-automated edge detection with manual control). Results depend on the ultrasound equipment in use, and moment of the cardiac cycle [113, 114]. cIMT varies between 5 - 10% or 0.03 mm, respectively, from systole to diastole and is thickest at end-diastolic moment [6, 22, 115]. It is therefore necessary, to provide appropriate reference values, assessed with a standardized protocol that is in line with the Mannheim Consensus [109] or the recently published statement from the Association for European Pediatric Cardiologists [69]. Furthermore, adherence to both, the standardized measurement protocol and ultrasound equipment, should be ensured [116]. Table 6 displays cIMT studies performed in children with no study that applied an identical examination protocol.

Table 6. Summary of ultrasound B-mode studies measuring intima-media thickness in children and adolescents. CCA = common carotid artery, ICA = internal carotid artery, and Bulb = carotid bulb.

Author	Subjects (females)	Age (years)	mean IMT (mm±SD) or IMT range (mm)	CCA	ICA	Bulb	Measurement location	Cardiac Cycle	Side left	Side right	Wall far	Wall near	Angle	IMT detection	IMT calculation
Aggoun [18]	n=57	11.1±3	0.5-0.53±0.03	x			1-2cm proximal the bifurcation, min. 1cm length	end-diastolic			x		N/A	automated	mean
Dawson [117]	n=635 (322)	11-34	0.49±0.04	x	x	x	N/A	N/A	x	x	x	x	3 diff. angles.	N/A	Mean
Iannuzzi [15]	n=100 (39)	6-14	0.51-0.54	x			1cm distance from the bifur-cation	N/A	x	x	x	x	N/A	N/A	max
Ishizu [67]	n=60 (33)	5-14	0.44±0.05	x			1-2cm proximal the bulb	end-diastolic	x	x	x		ant./ lat.	N/A	mean and max
Jarvisalo [16]	n=75 (27)	7-14	0.42-0.47±0.03	x		x	(a) 1-2cm proximal the bulb (b) left bulb region	end-diastolic	x	x	x		ant. oblique/ lateral	caliper	mean and max
Jarvisalo [8]	n=88 (33)	11±2	0.42-0.47±0.04	x			1-2cm proximal the bulb	end-diastolic	x	x	x		ant. oblique/ lateral	caliper	mean
Krantz [73]	n=229 (131)	12-25	0.54-0.56±0.06	x			distal CCA	N/A		x	x		N/A	automated	N/A
Krebs [38]	n=100 (53)	5-18	0.54	x			1cm proximal the bulb	N/A	x	x			lateral/ post.oblique	automated	mean and max
Lande [11]	n=56	10-18	0.53-0.93	x			1cm proximal the bifurcation	N/A	x		x		N/A	N/A	mean
Lavrencic [118]	n=56 (32)	11-27	0.49-0.71	x			(a) ICA, 1cm proximal (b) bifurcation (c) 1cm distal the bifurcation	N/A	x	x	x		N/A	N/A	max
Litwin [19]	n=110 (40)	6-20	0.41-0.45±0.05	x			1-2cm below the bifurcation, 1cm length	N/A	x	x	x		N/A	N/A	N/A
Menees [119]	n=49 (22)	6-19	0.46±0.04	x			2cm proximal the bifurcation, 1cm length	CRS/ non-CRS	x	x	x		N/A	semi-automated	mean

Author	Subjects (females)	Age (years)	mean IMT (mm±SD) or IMT range (mm)	CCA	ICA	Bulb	Measurement location	Cardiac Cycle	Side		Wall		Angle	IMT detection	IMT calculation
									left	right	far	near			
Meyer [13]	n=52 (29)	9-16	0.39±0.05 0.49±0.08	x		x	(a) 1cm distal the bulb (b) bifurcation	N/A	x	x	x	x	N/A	N/A	mean and max
Mittelman [120]	n=599 (307)	5-20	0.38±0.04	x			1cm proximal the bifurcation	N/A	x		x		N/A	automated	N/A
Sass [24]	n=193 (108)	10-24	0.48-0. 5±0.05	x			3cm proximal bifurcation, min. 1cm length	N/A	x	x	x		N/A	automated	mean
Böhm [25]	n=267 (143)	6-17	0.51	x			8-18mm proximal bifurcation, 1cm length	end-systolic		x	x		N/A	semi-automated	mean
Jourdan [22]	n=247 (127)	10-20	0.38-0.4±0.04	x			1-2cm proximal the bifurcation, 1cm length	N/A	x	x	x		N/A	caliper	N/A
Doyon [23]	n=1051	6-18	0.37-0.41	x			1-2cm proximal the bifurcation, 1cm length (Mannheim)	N/A	x	x	x		N/A	caliper or semi-automated	N/A
Pauciullo [72]	n=94 (45)	2-14	0.39-0.4±0.03	x			1cm proximal the bifurcation	systolic	x	x	x	x	ant./post.	N/A	mean and max
Reinehr [121]	n=124 (75)	9-13	0.4-0.6	x			near bifurcation	N/A	x	x	x		N/A	N/A	max
Singh [17]	n=66 (31)	10-18	0.32±0.08 0.33±0.05	x			1cm proximal the bulb, measuring 1,5cm proximal	N/A	x	x	x		N/A	N/A	N/A
Sorof [12]	n=32 (7)	13.9	0.64±0.12	x			2cm distal the flow divider	N/A	x	x	x		N/A	digital caliper	max
Tonstad [71]	n=90 (29)	10-19	0.48±0.07	x			(a) 1cm proximal the bulb (b) 1cm distal the bulb	end-diastolic		x	x		N/A	N/A	mean and max
Virkola [122]	n=46 (16)	2.7-19	0.33-0.5±0.1	x			2cm below the bifurcation	end-diastolic		x	x	x	N/A	electronic cali-per	N/A
Weberruß [70]	n=690 (380)	7-17	0.46±0.03	x			1cm proximal the bulb, 1cm length (Mannheim)	end-diastolic	x	x	x		N/A	semi-automated	mean
Woo [14]	n=72 (30)	7-12	0.45-0.49±0.04	x			1cm distal the bulb	N/A	x	x	x		ant/ lat./ post.oblique	automated	max

In a sub-sample of our cohort, we re-examined 30 cIMT measurements offline, to compare cIMT over a distance of 1 cm immediately after the bulb and over the same distance next to this first segment (Figure 10). According to Engelen et al. [123], the CCA segment of 0 to 2 cm proximal the bulb is the most common site of measurement, observed in a study cohort of 24.871 individuals, out of 24 study centers from 13 different countries. We observed no statistical differences between these two segments and concluded, that study results of comparable protocols that differed only in CCA location in-between this distance can be compared with each other [124].

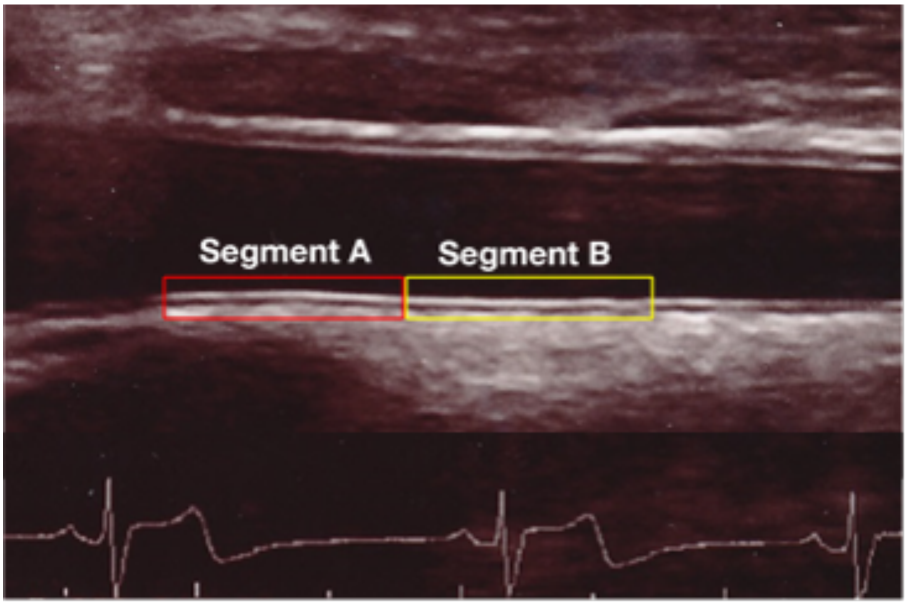

Figure 10. Comparison of two different carotid intima-media thickness measurement locations. Segment A refers to the distance of 1 cm immediately after the bulb, segment B is the distance over 1 cm next to segment A.

In this study, we examined cIMT at CCA far wall as in most pediatric studies [114], at end-diastolic moment according to the Mannheim Consensus [109]. cIMT was measured using a semi-automated edge-detection system with manual control, which is shown to provide the most accurate results [115]. Age-dependent cIMT reference values for boys and girls of this study correspond well with those by Ishizu et al. [67] but not with those recently assessed by Doyon et al. [23]. Divergence can be explained with aforementioned differences in cIMT assessment, as Doyon et al. [23] applied two different measurement methods (semi-automated and manual) and different examiners in seven study centers.

In the current literature, only Ishizu et al. [67] reported a moderate significant influence of age on cIMT ($r = 0.39$) controlled for other confounders. O'Leary and Bots [5] stated that in subjects younger than 18 years neither age nor sex significantly influenced cIMT. According to Cruickshank et al. [125] the change in cIMT between 8-17 years appears to be minimal, which was confirmed by results of this work.

Regarding sex-differences, non-significant results of our study for the overall study population and for most age groups are in accordance with others [22, 24, 67, 126]. Doyon et al. [23] reported significantly higher cIMT values in boys, starting at age 15 but not in younger age groups. Krebs et al. [127] investigated cIMT in $n = 270$ (126 girls) children and adolescents with type 1 diabetes, mean age 13.8 years, and reported significantly higher cIMT values in male than female patients (0.55 ± 0.07 vs. 0.57 ± 0.07, $p = .004$). The difference we observed was between boys and girls aged 8 - 10 years with boys having a higher cIMT than girls (0.46 ± 0.03 vs. 0.45 ± 0.03, $p = .005$).

One possible explanation could be a difference due to hormonal effects regarding an earlier onset of puberty within both sexes. As we did not assess pubertal status via Tanner stage this possible influence could only be hypothesized. However, pubertal hormones induce an earlier longitudinal growth in girls compared with boys [128]. One would expect that this earlier developmental process also affects other structures, resulting in higher instead of lower cIMT values in girls during puberty.

Only one study could be found, addressing a pubertal influence on cIMT, but in 10-18 year old type I diabetes patients [129], what limits a comparison between results of this study and our findings. The authors found no significant increase in cIMT during puberty in their sample consisting of 77 patients and 33 controls. As authors of this study did not investigate sex-differences, a possible pubertal influence cannot be ruled out [129].

Arterial stiffness is discussed to be the precursor of an increased IMT [18]. According to this, significant sex-differences in distensibility parameters would be expected to occur within the same age group than different cIMT values were observed (8-10 years). This presumption was not verified with our data (AC $p = .176$; Ep $p = .839$; β $p = .707$; PWV β $p = .925$).

Hidvegi et al. [130] noticed a steep increase in PWV values during puberty in a sample of 3374 boys and girls aged 3-18 years that started about 2 years earlier in girls compared to boys. Mean age, at which arterial stiffness in girls increased in this study was 10.1 years compared to an age span between 8-10 years in our sample. In contrast to this results [130] one study reported stiffer arteries in pre-pubertal girls compared to age-matched boys caused by an influence of sex steroid hormones on vessel structure [131]. If this effect would also influence cIMT values in pre-pubertal girls, it could be an explanation for sex-differences within our sub-sample. For further investigations regarding this matter, Tanner staging or assessment of hormonal status would be required. However, as long as possible sex-differences for both, cIMT and stiffness measures cannot be excluded so far, sex-dependent values should be reported.

Traditional cardiovascular risk factors, BMI and systolic BP, significantly influenced cIMT, as well as overweight and hypertensive subjects showed significantly increased cIMT values, which is in accordance to other studies [110, 132-135]. Within the obese sub-sample on the contrary, cIMT was not significantly different in obese boys and girls compared to sex- and age-matched normal weight controls. Furthermore, obese boys had nearly normal cIMT SDS (0.03). Higher cIMT values in obese girls (0.23) were still beyond the 75^{th} percentile (0.674). In multivariate regression analysis no significant influence of aforementioned risk factors (BMI and systolic BP) was observed in the obese sub-sample.

Whether or not obesity is an independent risk factor for an increased cIMT is under debate [136]. There are studies, reporting an increased cIMT in obese children [13, 14, 137, 138] while others observed the contrary [136, 139-141]. Di Salvo et al. [140] hypothesized that obesity as only risk factor does not contribute to increased cIMT values, which complies with our results. However, Morrison et al. [141] found no increased cIMT in children about the same age (5-16 years) and similar BMI SDS (2.3) than our sample, but with significantly increased systolic BP values, body fat and cholesterol levels as additional risk factors.

Aggoun et al. [139] and Tounian et al. [136] observed increased arterial stiffness, and endothelial dysfunction [136] in obese children but no significantly increased cIMT compared to lean controls. They hypothesized that increased arterial stiffness might not be sufficient to induce wall remodeling processes leading to increased cIMT values. We can only assume that the impact of obesity as only risk factor did not significantly influence cIMT, as other risk factors like cholesterol, glucose or triglyceride levels are lacking. Furthermore, it would have been important to record history of obesity and thus the duration of exposure to this risk factor [139]. Studies in adults verify a long-term effect with increased cIMT values in adults who had been overweight during childhood [142, 143].

Even though BMI is highly correlated with percentage of body fat (%BF), it does not regard weight distribution and distinguishes not between lean and fat mass, respectively [144, 145]. Thus alternative measures for BMI, as %BF, waist circumference (WC), waist-to-hip ratio (WHR) or waist-to-height ratio (WHtR) should also be considered in association to cIMT. In our study, WC and hip circumference were examined, to further calculate WHR and WHtR. All parameters were significantly higher in obese compared to normal weight subjects ($p < .001$), whereas no measure significantly correlated with cIMT (WC $p = .641$, WHR $p = .303$, WHtR $p = .742$). Aggoun et al. [139] and Morrison et al. [141] investigated the influence of %BF and WC on cIMT. They found no significant association with %BF [139, 141], and only a small association with WC ($R^2 = 0.03$, $p = .004$) [141]. These results support our findings and emphasize a long-term follow-up to investigate long-term effects of overweight and obesity on vascular parameters during childhood and adolescence [142].

4.2 Arterial Distensibility: Influence of BMI and Differences in Obese Girls but not Boys

Arterial distensibility and compliance or arterial stiffness are terms, being used interchangeably in the current literature. However, they address different aspects of the artery studied [7, 146]. Arterial compliance is the absolute change in diameter for a given increase in pressure [111] and specifies the vessel's ability to store volume and reduce the pressure increase during systole [147]. Distensibility is the relative change in diameter for a given pressure change [7]. Arterial stiffness can be defined as resistance to BP induced deformation [148] or rigidity of the arterial wall [7]. In this study, the term distensibility was used as umbrella term for arterial compliance, elastic modulus, stiffness index β, and local pulse wave velocity β.

The higher arterial compliance, the more elastic is an artery, the higher Ep, β, and PWV β on the contrary, the stiffer is an artery. As well as cIMT, these parameters also depend strongly on the measurement protocol and measurement device they were obtained. Arterial distensibility can be measured by applanation tonometry, oscillometric technique, MRI, and ultrasound [149]. Carotid-femoral (cf) PWV is the gold standard to assess arterial stiffness [150, 151]. Generally, PWV provides information about regional stiffness, whereas AC, Ep, and β refer to local stiffness of the vessel studied [7, 149]. In this study, arterial distensibility was assessed via ultrasound with radio frequency echo tracking (ProSound Alpha 6; Aloka/ Hitachi Medical Systems GmbH, Wiesbaden, Germany), which measures indices locally at the carotid artery. PWV is therefore defined as local PWV derived from β (PWV β).

Reference values for distensibility parameters assessed with the Aloka/ Hitachi ProSound system only exist for the Chinese population as unpublished producer information without proper description of the respective methodology (Figure 11). Comparison between Chinese reference values with our data is displayed in Figure 12 for children younger than 10 years and children in between 10-17 years (our data) and 10-19 years (Chinese data), respectively. However, the rationale behind comparing those two cohorts is questionable as the measuring protocol for the Chinese sample was not accessible.

Among distensibility parameters in our reference population AC significantly correlated with age ($r = -0.18$, $p < .001$) indicating less elastic arteries with increasing age [111, 152]. Arterial stiffness is the reciprocal of compliance, hence it seems rational that Ep, β, and PWV β were inversely correlated with AC (Ep $r = -0.65$, β $r = -0.63$, and PWV β $r = -0.68$, $p < .001$).

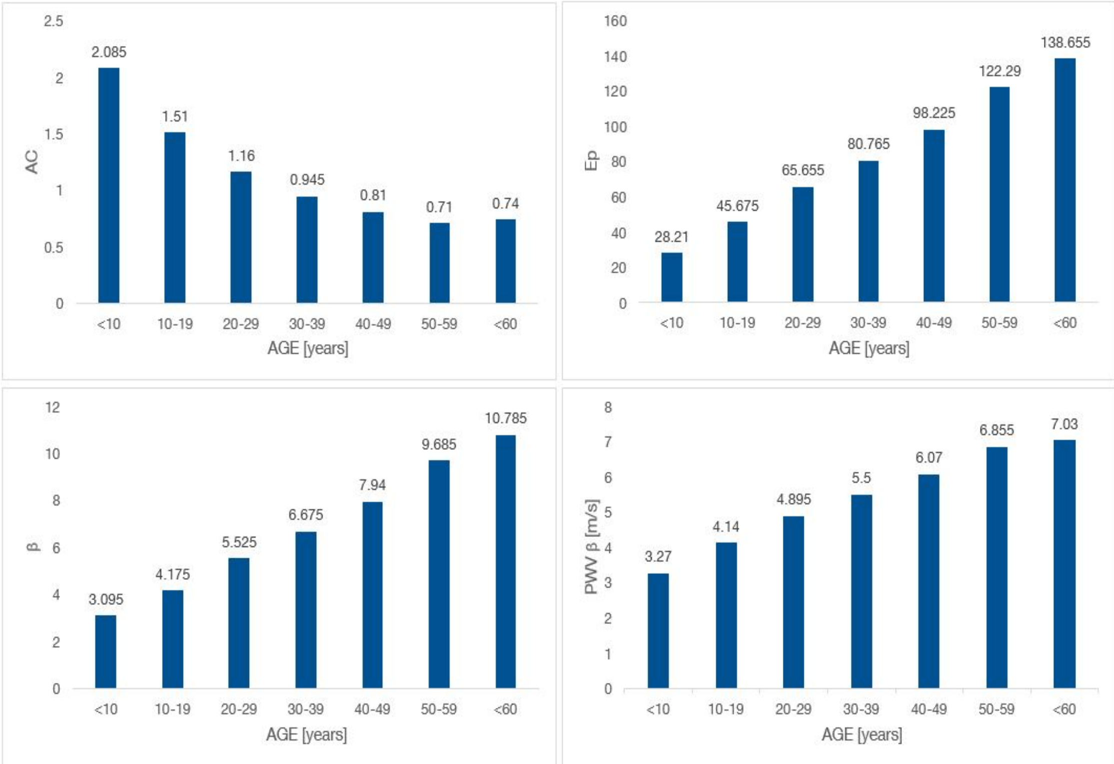

Figure 11. Chinese reference values for arterial compliance (AC), elastic modulus (Ep), stiffness index β (β), and local pulse wave velocity β (PWV β) assessed in 1 971 men and 2 841 women of different age groups.

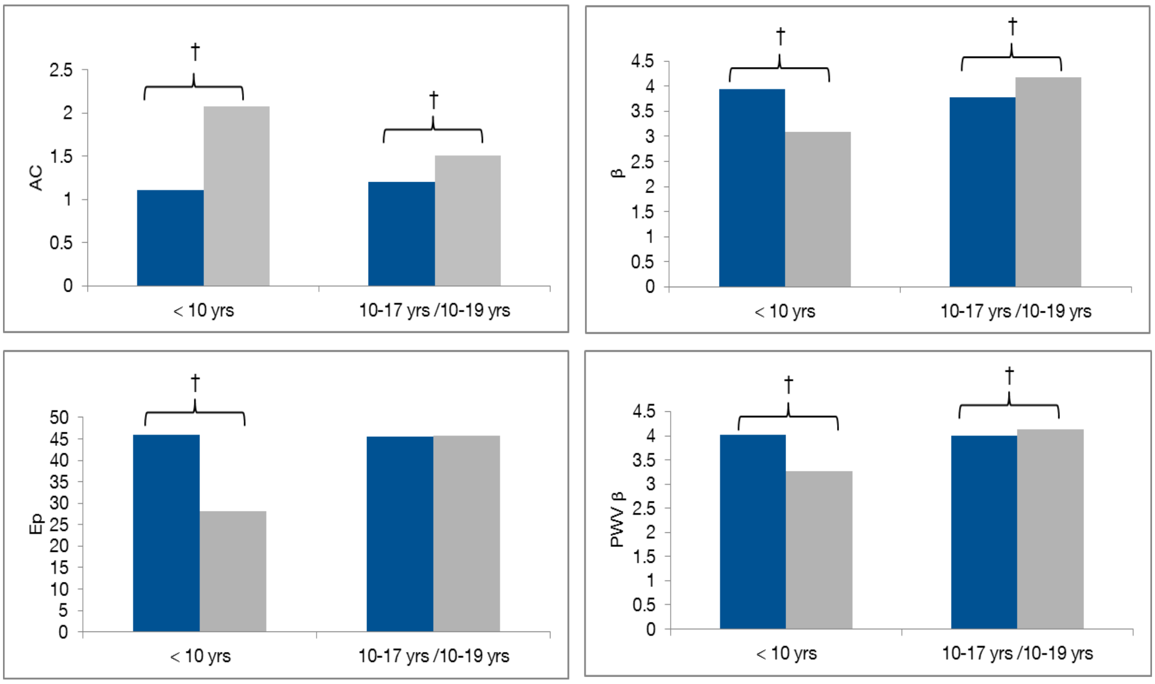

Figure 12. Comparison of distensibility parameters arterial compliance (AC), elastic modulus (Ep), stiffness index β (β), and local pulse wave velocity β (PWV β) between the present study (blue columns) and Chinese reference values (grey columns). † $p < 0.001$.

Most studies address either parameters of compliance or stiffness [153-156] and do not compare these indices with each other. Jourdan et al. [22] and Doyon et al. [23] investigated compliance and stiffness, with both being affected by age, BMI, and BP. There's discrepancy in our results, with AC being significantly influenced by BMI, whereas stiffness indices are not.

According to the current literature [154, 157-159], a higher BMI was associated with higher AC as well as AC of our obese sub-group was higher compared to normal weight controls. The underlying mechanisms are hypothesized to be linked to accelerated pubertal development in overweight and obese children that increases arterial compliance, as well as chronic vasodilation, caused by larger blood volume in overweight subjects [153, 160].

Why stiffness parameters were not significantly affected by BMI is not clear, as they were assessed within the same measurement as AC. All parameters are derived from the same dependent variables, which are systolic and diastolic BP as well as end-systolic and end-diastolic vessel diameter. AC and stiffness parameters are differently affected by BP [22] what may be one explanation. The other might be mechanisms acting on a molecular, cellular, and genetic level, which is far beyond the scope of this study [161].

Within a healthy vessel, there is an equilibrium between collagen, responsible for wall stability, and elastin, responsible for elasticity. If this equilibrium is being disturbed, by either endogenous causes (inflammation) or mechanical stress (hypertension), an increase in collagen or destruction of elastin components might be the consequence [162]. This may affect arterial compliance and arterial stiffness to a different extent.

There is no agreement upon increased systolic BP and pulse pressure (PP) are precursors or successors of impaired distensibility [147, 163]. We have observed a significant inverse correlation of systolic BP with AC and positive correlation with stiffness parameters. Diastolic BP was positively associated with AC and negatively with stiffness parameters. These results are in accordance with identification of a high PP being associated with stiffer arteries [111].

In contrast to the positive association between BMI and AC in our reference population, there was no significant difference in AC between the obese sub-sample and normal weight controls. However, obese girls had significantly increased Ep, β, and PWV β SDS, compared with normal weight girls. There are two aspects to address in this matter. Firstly, the association of obesity and arterial stiffness, and secondly, the interaction between sex and obesity as significantly increased stiffness parameters were only observed in girls. The hormone leptin, which is produced by adipocytes, is discussed as mediator of arterial stiffness in obese subjects [164, 165]. This effect might be limited to obese subjects only and not be detected in the overweight status, as we did not observe a significant influence of BMI on arterial stiffness in our reference population. Cote et al. [146] reviewed 15 studies on obesity and arterial stiffness in children with only two studies, that observed higher arterial stiffness in children with a normal BMI compared to obese children [158, 166].

There could be an interaction between sex and obesity, as stiffer arteries were only observed in girls but not in boys. This interaction might be modulated by hormonal levels and an earlier onset of puberty in girls, leading to increased arterial stiffness in obese girls [130]. Ahimastos et al. [131] on the contrary, report stiffer arteries in pre-pubertal girls (10.3 ± 0.1 years) compared with boys (10.3 ± 0.1 years), but not in post-pubertal girls compared with boys (15.9 ± 0.2 and 15.9 ± 0.4 years). We can only assume this influence of obesity on arterial stiffness but cannot capture what happens during puberty. Furthermore, we do not know the time span, pathological adaptation takes until obesity results in stiffer arteries and how obesity interacts with age, sex, and pubertal status [146].

4.3 Health-Related Fitness: Influence of Fitness and Strength on Carotid Intima-Media Thickness and Arterial Distensibility

This is the first study, investigating cardiovascular data (cIMT and arterial distensibility) and health-related fitness (FITNESSGRAM®) in a large sample of n = 697 German boys and girls, aged 7-17 years. Authors hypothesized, that cardiorespiratory fitness is positively associated with healthier arteries, regarding a thinner cIMT, a higher AC, and decreased Ep, β, and PWV β in fit subjects. Strength was furthermore expected to have no negative influence on arterial parameters.

Regarding cardiorespiratory fitness, the hypothesis could only be confirmed for distensibility parameters. cIMT however was positively correlated with cardiorespiratory fitness, indicating a thicker cIMT with increasing fitness. As distensibility parameters were positively affected by cardiorespiratory fitness, shown by increased AC and reduced arterial stiffness, we state a functional adaptation of the arterial wall, similar to adaptations in the athlete's heart [167]. Giannattasio et al. [168] assume that exercise leads to an increase in cIMT in adults, which involves rather distensible tissues than less elastic collagen, and preserves a healthy vessel function.

It is not clear, if physical fitness alters IMT in younger subjects and in which direction [169]. Comparing boys and girls of different fitness levels (low, moderate, high), there were no significant differences between categories in accordance to Pahkala et al. [90]. However, they observed the abdominal aorta, too, and found significantly lower IMT values between boys and girls with a high fitness level compared to moderate and low levels. The authors concluded, that arterial alterations may first start in the aorta and will later be detectable at the carotid artery [90]. No association between fitness and cIMT either is reported by Ried-Larsen et al. [91]. Ferreira et al. [95] found no significant decreased cIMT in adults with high levels of cardiorespiratory fitness during adolescence. In obese children however, who already had significantly increased cIMT values compared to normal weight controls, an increase in physical activity and physical fitness, is associated with a reduction in cIMT [170, 171].

Regarding arterial distensibility, cardiorespiratory fitness was revealed to have an independent positive influence on distensibility parameters in multivariate regression analysis, which is in accordance with other studies in children [90, 91, 97, 98] and adults [96, 98, 156, 172, 173].

The mechanisms leading to improved arterial distensibility are discussed to be caused by increased shear stress and nitric oxide release, which positively modifies vasodilatory response of the vessel wall, lowers BP and heart rate, and positively modifies further cardiovascular risk factors [98, 172, 174]. It is beyond the scope of this work to explore these mechanisms, which cannot be further investigated with the data assessed.

In this study, fitness was inversely correlated with BMI SDS ($r = -0.298$, $p < .001$), WHtR ($r = -0.385$, $p < .001$), and WHR ($r = -0.37$, $p < .001$). According to the fat-but-fit hypotheses in adults [175], Jago et al. [87] and Eisenmann et al. [176] proved high fitness levels being superior to BMI in relation to cardiovascular risk. The inverse correlation of cardiorespiratory fitness with BMI SDS, WHtR, and WHR, shows that fitness favorably modifies body composition and therefore reduces cardiovascular risk [169].

Contrary to aerobic exercise and arterial distensibility, findings in adults about strength or resistance training to improve strength, respectively, are contradictory. Results depend on subject’s age and basal BP levels, training intensity and type of contraction (eccentric or concentric), muscles or muscle groups involved, and if central or peripheral distensibility is assessed [172, 177, 178]. In children no study was found in pubmed and medline databases by searching for “strength”, “resistance training”, “distensibility”, “arterial stiffness”, “children”, and “adolescents”.

To apply results observed in adults is difficult, as resistance training in children and adolescents may vary in comparison with adults [179]. Furthermore, we did not investigate effects of a resistance training intervention [178, 180] and assessed muscular endurance and not maximum strength with two FITNESSGRAM® tasks (push-ups and curl-ups), whereas studies in adults used strength training devices [181-183].

In our sample, strength was no independent predictor of cIMT or arterial distensibility, and none of the strength tasks (push-ups and curl-ups) was significantly correlated with parameters of arterial distensibility. However, strength parameters were positively correlated with cardiorespiratory fitness (push-ups: $r = 0.584$, $p < .001$, curl-ups: $r = 0.386$, $p < .001$). Push-ups and curl-ups were intercorrelated ($r = 0.407$, $p < .001$), too, as well as both parameters showed an inverse correlation with BMI SDS ($r = -0.257$ for push-ups, $p < .001$ and $r = -0.132$ for curl-ups), WHtR ($r = -0.312$ and $r = -0.21$, $p < .001$), and WHR ($r = -0.244$ and $r = -0.208$, $p < .001$).

These results indicate that children, with a good cardiorespiratory fitness achieve good results in strength testing, too, and positively influence their body composition. Thus, the essential finding of our study is, that cardiorespiratory fitness has a positive effect on vascular health, already in children and adolescents, and that muscular strength does not impair vascular function.

5 LIMITATIONS OF THE STUDY

A bias in selection of study participants cannot be ruled out. Out of 41 schools of the district "Berchtesgadener Land", 12 schools decided to participate in the project. Grade and number of classes per school were determined by the principal. Study participation was voluntary after written informed consent form was signed by parents (of children < 14 years) and parents and children (> 14 years). Parents and/ or students worried about negative study results might have not agreed to participate and vice versa, parents and/ or students who are interested in their health and lead a healthy lifestyle might have joined the project. Attendance or non-attendance rate, respectively, was not assessed to rebut this selection bias. A further bias exists, as data was collected in one school district in southern Bavaria and not in different schools of different districts.

Due to technical problems with the ultrasound storage function, 281 cIMT data sets and 89 distensibility data sets were lost. This diminished the study population especially within younger age groups.

The proportion of children and adolescents with a systolic or diastolic BP measurement or both above the 95th percentile was higher than in the average German population. Within the entire sample, 28.2% (147 boys/ 132 girls) had systolic BP values > 95th percentile, 16.2% diastolic BP (72/ 89), and 10.4% (49/ 54) both, systolic and diastolic BP values > 95th percentile. Due to a relatively tight schedule, only one BP measurement was taken and not two according to German guidelines [184]. We defined children with BP values > 95th not as hypertensive but with "suspected hypertension".

BP was a major determinant of parameters of arterial distensibility that could statistically not be controlled. The informative value of our results in relation to BP must therefore be interpreted carefully and according to BP levels. Prevalence of hypertension was higher in younger age groups, who might have been more excited or more nervous than older subjects, similar to the white coat effect. To increase precision of data acquisition would have taken more time and more personnel requirements - or would have resulted in less subjects examined.

The assessment of pubertal status via Tanner staging would have strengthened our results as well as measuring blood cholesterol, triglycerides and glucose levels. On the other hand, a positive ethic vote would have been difficult with venous blood sampling and participation rates might have dropped. Whereas history of obesity and percentage of body fat would have been easy to assess and could help the interpretation regarding the influence of BMI on vascular data.

To test health-related fitness, a 6 or 12 minute run test would have been a validated alternative to the 20 meter shuttle-run applied in this study. We chose the shuttle-run however, as the testing facility had no gym (6 minute run test) and testing was also during autumn and winter season (12 minute run test). For strength testing maximum handgrip test would have been an easy alternative to assume for whole body strength in addition to FITNESSGRAM®, as it can easily be assessed in healthy subjects and patient collectives, too.

Arterial distensibility was assessed with an ultrasound device, for which no comparable data existed. This made interpretation of distensibility parameters applied in this study rather difficult.

The added value of cIMT is furthermore being discussed, in addition to existing risk scores [185], in adult patient groups [186], or compared to other subclinical markers [187]. There is to reply, that in children, where atherosclerotic plaques and cardiovascular events are unlikely to occur, cIMT provides an easy tool to investigate pre-atherosclerotic changes. In addition with parameters of arterial distensibility we established reference values that can be applied in patient collectives or longitudinal studies of the same sample.

6 CONCLUSIONS

This study investigated cIMT and arterial distensibility following a highly standardized protocol according to the Mannheim Consensus [109] in the to-date largest German cohort of boys and girls, aged 7-17 years. Reference values for cIMT and arterial distensibility were published [70] to be applied in future studies in healthy children and pediatric patient groups.

Traditional cardiovascular risk factors BMI and BP significantly influenced vascular parameters in the reference population, whereas inconsistent results were observed within the obese sub-sample. It was beyond the scope of this work, to investigate specific mechanisms, leading to different alterations of the arterial wall in different samples. Consistent with the literature, we hypothesize and interaction of genetic, hormonal, metabolic, and lifestyle factors that contribute to atherogenesis [57].

A positive effect of cardiorespiratory fitness on vascular parameters could be detected within this study. Underlying reasons might be manifold, too, such as reduced mechanical stress (lower BP and heart rate), positive influence of fitness on metabolic parameters (e.g. blood levels, insulin resistance), improved body composition, and a healthy lifestyle (non-smoking, healthy diet). Reduced physical fitness is revealed as independent risk factor for cardiovascular disease [84] and associated with higher prevalence of overweight and obesity in children [188]. Furthermore, childhood BMI is positively correlated with adult BMI levels [189]. Fitness levels in childhood on the other hand, are inversely correlated with cardiovascular risk in adulthood [88, 89, 190].

Physical fitness in this study was revealed as very important aspect of cardiovascular risk prevention. It positively affected arterial parameters in children, and may prevent atherosclerotic processes to onset during childhood and have a beneficial long-term effect on future cardiovascular health.

7 REFERENCES

1. Strong, J.P. and G.H. Mc, Jr., *The natural history of coronary atherosclerosis.* Am J Pathol, 1962. **40**: p. 37-49.
2. Strong, J.P., *The natural history of atherosclerosis in childhood.* Ann N Y Acad Sci, 1991. **623**: p. 9-15.
3. Urbina, E.M., et al., *Noninvasive assessment of subclinical atherosclerosis in children and adolescents: recommendations for standard assessment for clinical research: a scientific statement from the American Heart Association.* Hypertension, 2009. **54**(5): p. 919-50.
4. WHO, *Global Atlas on Cardiovascular Disease Prevention Control*, W.H. Organization, Editor. 2011: Geneva.
5. O'Leary, D.H. and M.L. Bots, *Imaging of atherosclerosis: carotid intima-media thickness.* Eur Heart J, 2010. **31**(14): p. 1682-9.
6. O'Leary, D.H., et al., *Carotid-artery intima and media thickness as a risk factor for myocardial infarction and stroke in older adults. Cardiovascular Health Study Collaborative Research Group.* N Engl J Med, 1999. **340**(1): p. 14-22.
7. Mackenzie, I.S., I.B. Wilkinson, and J.R. Cockcroft, *Assessment of arterial stiffness in clinical practice.* QJM, 2002. **95**(2): p. 67-74.
8. Jarvisalo, M.J., et al., *Increased aortic intima-media thickness: a marker of preclinical atherosclerosis in high-risk children.* Circulation, 2001. **104**(24): p. 2943-7.
9. Wiegman, A., et al., *Arterial intima-media thickness in children heterozygous for familial hypercholesterolaemia.* The Lancet, 2004. **363**(9406): p. 369-370.
10. Koeijvoets, K.C., et al., *Low-density lipoprotein receptor genotype and response to pravastatin in children with familial hypercholesterolemia: substudy of an intima-media thickness trial.* Circulation, 2005. **112**(20): p. 3168-73.
11. Lande, M.B., et al., *Effects of childhood primary hypertension on carotid intima media thickness: a matched controlled study.* Hypertension, 2006. **48**(1): p. 40-4.
12. Sorof, J.M., et al., *Carotid Artery Intimal-Medial Thickness and Left Ventricular Hypertrophy in Children With Elevated Blood Pressure.* Pediatrics, 2003. **111**(1): p. 61-66.
13. Meyer, A.A., et al., *Impaired flow-mediated vasodilation, carotid artery intima-media thickening, and elevated endothelial plasma markers in obese children: the impact of cardiovascular risk factors.* Pediatrics, 2006. **117**(5): p. 1560-7.

14. Woo, K.S., et al., *Overweight in children is associated with arterial endothelial dysfunction and intima-media thickening.* Int J Obes Relat Metab Disord, 2004. **28**(7): p. 852-7.
15. Iannuzzi, A., et al., *Carotid artery stiffness in obese children with the metabolic syndrome.* Am J Cardiol, 2006. **97**(4): p. 528-31.
16. Jarvisalo, M.J., et al., *Endothelial dysfunction and increased arterial intima-media thickness in children with type 1 diabetes.* Circulation, 2004. **109**(14): p. 1750-5.
17. Singh, T.P., H. Groehn, and A. Kazmers, *Vascular function and carotid intimal-medial thickness in children with insulin-dependent diabetes mellitus.* Journal of the American College of Cardiology, 2003. **41**(4): p. 661-665.
18. Aggoun, Y., et al., *Arterial Mechanical Changes in Children With Familial Hypercholesterolemia.* Arteriosclerosis, Thrombosis, and Vascular Biology, 2000. **20**(9): p. 2070-2075.
19. Litwin, M., et al., *Intima-media thickness and arterial elasticity in hypertensive children: controlled study.* Pediatr Nephrol, 2004. **19**(7): p. 767-74.
20. Giannattasio, C., et al., *Early impairment of large artery structure and function in type I diabetes mellitus.* Diabetologia, 1999. **42**(8): p. 987-94.
21. Iannuzzi, A., et al., *Increased carotid intima-media thickness and stiffness in obese children.* Diabetes Care, 2004. **27**(10): p. 2506-8.
22. Jourdan, C., et al., *Normative values for intima-media thickness and distensibility of large arteries in healthy adolescents.* J Hypertens, 2005. **23**(9): p. 1707-15.
23. Doyon, A., et al., *Carotid artery intima-media thickness and distensibility in children and adolescents: reference values and role of body dimensions.* Hypertension, 2013. **62**(3): p. 550-6.
24. Sass, C., et al., *Intima-media thickness and diameter of carotid and femoral arteries in children, adolescents and adults from the Stanislas cohort: effect of age, sex, anthropometry and blood pressure.* J Hypertens, 1998. **16**(11): p. 1593-602.
25. Bohm, B., et al., *Sex differences of carotid intima-media thickness in healthy children and adolescents.* Atherosclerosis, 2009. **206**(2): p. 458-63.
26. Lorenz, M.W., et al., *Prediction of clinical cardiovascular events with carotid intima-media thickness: a systematic review and meta-analysis.* Circulation, 2007. **115**(4): p. 459-67.

27. Schmermund, A., *[Cardiorespiratory fitness. Importance of sports for health].* Herz, 2004. **29**(4): p. 365-72.
28. Franklin, B.A. and P.A. McCullough, *Cardiorespiratory fitness: an independent and additive marker of risk stratification and health outcomes.* Mayo Clin Proc, 2009. **84**(9): p. 776-9.
29. Green, D.J., et al., *Exercise and cardiovascular risk reduction: time to update the rationale for exercise?* J Appl Physiol (1985), 2008. **105**(2): p. 766-8.
30. Sakamoto, H. and E. Murakami, *Atherosclerosis: Risk Factors, Prevention, and Treatment.* 2012, New York: Nova Science Publishers, Inc. .
31. Anderson, P.G. and J.B. Atkinson, *Atherosclerosis*, in *Atlas of cardiovascular pathology for the clinician*, B. McManus, Editor. 2008, Current Medicine Group: Philadelphia. p. 68-84.
32. Ware, R.W., *Does Cholesterol Drive Coronary Atherosclerosis?*, in *Atherosclerosis: Risk Factors, Prevention, and Treatment*, H. Sakamoto and E. Murakami, Editors. 2012, New Science Publishers: New York. p. 1-40.
33. Holman, R.L., et al., *The natural history of atherosclerosis: the early aortic lesions as seen in New Orleans in the middle of the of the 20th century.* Am J Pathol, 1958. **34**(2): p. 209-35.
34. Stary, H.C., *Evolution and progression of atherosclerotic lesions in coronary arteries of children and young adults.* Arteriosclerosis, 1989. **9**(1 Suppl): p. I19-32.
35. Newman, W.P., 3rd, W. Wattigney, and G.S. Berenson, *Autopsy studies in United States children and adolescents. Relationship of risk factors to atherosclerotic lesions.* Ann N Y Acad Sci, 1991. **623**: p. 16-25.
36. Fowler, S.D., et al., *Cellular models of atherosclerosis in the young.* Ann N Y Acad Sci, 1991. **623**: p. 60-9.
37. Dreßler, H.T. and K.P. Göttmann, *Pathophysiologie der koronaren Herzkrankheit*, in *Koronare Herzkrankheit*, H.A. Dietrich and H. Mörl, Editors. 1993, WVG mbH: Stuttgart. p. 29-42.
38. Krebs, A., et al., *Synergistic effects of elevated systolic blood pressure and hypercholesterolemia on carotid intima-media thickness in children and adolescents.* Pediatr Cardiol, 2009. **30**(8): p. 1131-6.
39. Ross, R., J. Glomset, and L. Harker, *Response to injury and atherogenesis.* Am J Pathol, 1977. **86**(3): p. 675-84.
40. Goldstein, J.L. and M.S. Brown, *Hyperlipidemia in coronary heart disease: a biochemical genetic approach.* J Lab Clin Med, 1975. **85**(1): p. 15-25.

41. Steiner, I. and J. Laco, *[Rokitansky on atherosclerosis].* Cesk Patol, 2008. **44**(1): p. 23-4.
42. Mayerl, C., et al., *Atherosclerosis research from past to present--on the track of two pathologists with opposing views, Carl von Rokitansky and Rudolf Virchow.* Virchows Arch, 2006. **449**(1): p. 96-103.
43. Sitia, S., et al., *From endothelial dysfunction to atherosclerosis.* Autoimmun Rev, 2010. **9**(12): p. 830-4.
44. McGill, H.C.J., et al., *Origin of atherosclerosis in childhood and adolescence.* Am J Clin Nutr, 2000. **72**((suppl)): p. 1307S-15S.
45. McGill, H.C., Jr., et al., *Origin of atherosclerosis in childhood and adolescence.* Am J Clin Nutr, 2000. **72**(5 Suppl): p. 1307S-1315S.
46. Tracy, R.E., et al., *Risk factors and atherosclerosis in youth autopsy findings of the Bogalusa Heart Study.* Am J Med Sci, 1995. **310 Suppl 1**: p. S37-41.
47. O'Donnell, C.J. and R. Elosua, *[Cardiovascular risk factors. Insights from Framingham Heart Study].* Rev Esp Cardiol, 2008. **61**(3): p. 299-310.
48. Juonala, M., J.S. Viikari, and O.T. Raitakari, *Main findings from the prospective Cardiovascular Risk in Young Finns Study.* Curr Opin Lipidol, 2013. **24**(1): p. 57-64.
49. Anderson, K.M., et al., *An updated coronary risk profile. A statement for health professionals.* Circulation, 1991. **83**(1): p. 356-62.
50. Assmann, G., P. Cullen, and H. Schulte, *Simple scoring scheme for calculating the risk of acute coronary events based on the 10-year follow-up of the prospective cardiovascular Munster (PROCAM) study.* Circulation, 2002. **105**(3): p. 310-5.
51. Grundy, S.M., et al., *Implications of recent clinical trials for the National Cholesterol Education Program Adult Treatment Panel III guidelines.* Circulation, 2004. **110**(2): p. 227-39.
52. Hu, G., et al., *The joint associations of occupational, commuting, and leisure-time physical activity, and the Framingham risk score on the 10-year risk of coronary heart disease.* Eur Heart J, 2007. **28**(4): p. 492-8.
53. Maahs, D.M., et al., *Cardiovascular disease risk factors in youth with diabetes mellitus: a scientific statement from the American Heart Association.* Circulation, 2014. **130**(17): p. 1532-58.
54. Wilson, P.W., et al., *Prediction of coronary heart disease using risk factor categories.* Circulation, 1998. **97**(18): p. 1837-47.
55. van Rooy, M.J. and E. Pretorius, *Obesity, hypertension and hypercholesterolemia as risk factors for atherosclerosis*

leading to ischemic events. Curr Med Chem, 2014. **21**(19): p. 2121-9.
56. Berenson, G.S., et al., *Association between multiple cardiovascular risk factors and atherosclerosis in children and young adults. The Bogalusa Heart Study.* N Engl J Med, 1998. **338**(23): p. 1650-6.
57. Novo, S., et al., *Risk Factors and Atherosclerotic Disease*, in *Preclinical Atherosclerosis, Global Cardiovascular Risk and Cardiovascular Events*, S. Novo, Editor. 2012, Bentham Science Publishers. p. 7-21.
58. Packard, R.R. and P. Libby, *Inflammation in atherosclerosis: from vascular biology to biomarker discovery and risk prediction.* Clin Chem, 2008. **54**(1): p. 24-38.
59. Salonen, J.T. and R. Salonen, *Ultrasonographically assessed carotid morphology and the risk of coronary heart disease.* Arteriosclerosis, Thrombosis, and Vascular Biology, 1991. **11**(5): p. 1245-1249.
60. Bots, M.L., et al., *Common carotid intima-media thickness and risk of stroke and myocardial infarction: the Rotterdam Study.* Circulation, 1997. **96**(5): p. 1432-7.
61. Pignoli, P., et al., *Intimal plus medial thickness of the arterial wall: a direct measurement with ultrasound imaging.* Circulation, 1986. **74**(6): p. 1399-406.
62. Bots, M.L., et al., *Cross-sectionally assessed carotid intima-media thickness relates to long-term risk of stroke, coronary heart disease and death as estimated by available risk functions.* J Intern Med, 1999. **245**(3): p. 269-76.
63. Bauer, M., et al., *Carotid intima-media thickness as a biomarker of subclinical atherosclerosis.* Swiss Med Wkly, 2012. **142**: p. w13705.
64. Ludwig, M., et al., *[Intima media thickness of the carotid arteries: early pointer to arteriosclerosis and therapeutic endpoint].* Ultraschall Med, 2003. **24**(3): p. 162-74.
65. Howard, G., et al., *Carotid artery intimal-medial thickness distribution in general populations as evaluated by B-mode ultrasound. ARIC Investigators.* Stroke, 1993. **24**(9): p. 1297-1304.
66. Stein, J.H., et al., *Use of carotid ultrasound to identify subclinical vascular disease and evaluate cardiovascular disease risk: a consensus statement from the American Society of Echocardiography Carotid Intima-Media Thickness Task Force. Endorsed by the Society for Vascular Medicine.* J Am Soc Echocardiogr, 2008. **21**(2): p. 93-111; quiz 189-90.

67. Ishizu, T., et al., *Effect of age on carotid arterial intima-media thickness in childhood.* Heart Vessels, 2004. **19**(4): p. 189-95.
68. Bauer, M., et al., *The effect of age and risk factors on coronary and carotid artery atherosclerotic burden in males-Results of the Heinz Nixdorf Recall Study.* Atherosclerosis, 2009. **205**(2): p. 595-602.
69. Dalla Pozza, R., et al., *Intima media thickness measurement in children: A statement from the Association for European Paediatric Cardiology (AEPC) Working Group on Cardiovascular Prevention endorsed by the Association for European Paediatric Cardiology.* Atherosclerosis, 2015. **238**(2): p. 380-387.
70. Weberruss, H., et al., *Increased intima-media thickness is not associated with stiffer arteries in children.* Atherosclerosis, 2015. **242**(1): p. 48-55.
71. Tonstad, S., et al., *Risk factors related to carotid intima-media thickness and plaque in children with familial hypercholesterolemia and control subjects.* Arterioscler Thromb Vasc Biol, 1996. **16**(8): p. 984-91.
72. Pauciullo, P., et al., *Increased intima-media thickness of the common carotid artery in hypercholesterolemic children.* Arterioscler Thromb, 1994. **14**(7): p. 1075-9.
73. Krantz, J.S., et al., *Early onset of subclinical atherosclerosis in young persons with type 1 diabetes.* J Pediatr, 2004. **145**(4): p. 452-7.
74. Mattsson, N., et al., *Arterial structure and function in young adults with the metabolic syndrome: the Cardiovascular Risk in Young Finns Study.* Eur Heart J, 2008. **29**(6): p. 784-91.
75. Ross, R., *Atherosclerosis is an inflammatory disease.* Am Heart J, 1999. **138**(5 Pt 2): p. S419-20.
76. Bots, M.L., et al., *Carotid intima-media thickness, arterial stiffness and risk of cardiovascular disease: current evidence.* J Hypertens, 2002. **20**(12): p. 2317-25.
77. Nichols, W.W., M.F. O'Rourke, and C. Vlachopoulos, *McDonald's Blood Flow in Arteries*. 6th Edition ed. 2011, Boca Raton, FL: Taylor & Francis Group.
78. O'Rourke, M. and E.D. Frohlich, *Pulse pressure: Is this a clinically useful risk factor?* Hypertension, 1999. **34**(3): p. 372-4.
79. Haller, M.J., et al., *Radial artery tonometry demonstrates arterial stiffness in children with type 1 diabetes.* Diabetes Care, 2004. **27**(12): p. 2911-7.

80. Boutouyrie, P., et al., *Aortic stiffness is an independent predictor of primary coronary events in hypertensive patients: a longitudinal study.* Hypertension, 2002. **39**(1): p. 10-5.
81. Aloka, *Aloka Prosound a7. How to use.*, A. co.ltd., Editor. 2008.
82. Cooper and Insitute, *Fitnessgram & Activitygram. Test Administration Manual.*, C. Insitute, Editor. 2010, Human Kinetics: Dallas, Texas.
83. Caspersen, C.J., K.E. Powell, and G.M. Christenson, *Physical activity, exercise, and physical fitness: definitions and distinctions for health-related research.* Public Health Rep, 1985. **100**(2): p. 126-31.
84. Kodama, S., et al., *Cardiorespiratory fitness as a quantitative predictor of all-cause mortality and cardiovascular events in healthy men and women: a meta-analysis.* JAMA, 2009. **301**(19): p. 2024-35.
85. Seals, D.R., et al., *Habitual exercise and arterial aging.* J Appl Physiol (1985), 2008. **105**(4): p. 1323-32.
86. Liu, J., et al., *Effects of cardiorespiratory fitness on blood pressure trajectory with aging in a cohort of healthy men.* J Am Coll Cardiol, 2014. **64**(12): p. 1245-53.
87. Jago, R., et al., *Fatness, fitness, and cardiometabolic risk factors among sixth-grade youth.* Med Sci Sports Exerc, 2010. **42**(8): p. 1502-10.
88. Carnethon, M.R., et al., *Cardiorespiratory fitness in young adulthood and the development of cardiovascular disease risk factors.* JAMA, 2003. **290**(23): p. 3092-100.
89. Ruiz, J.R., et al., *Predictive validity of health-related fitness in youth: a systematic review.* Br J Sports Med, 2009. **43**(12): p. 909-23.
90. Pahkala, K., et al., *Association of fitness with vascular intima-media thickness and elasticity in adolescence.* Pediatrics, 2013. **132**(1): p. e77-84.
91. Ried-Larsen, M., et al., *Physical activity intensity and subclinical atherosclerosis in Danish adolescents: the European Youth Heart Study.* Scand J Med Sci Sports, 2013. **23**(3): p. e168-77.
92. Popovic, M., et al., *The effects of endurance and recreational exercise on subclinical evidence of atherosclerosis in young adults.* Am J Med Sci, 2010. **339**(4): p. 332-6.
93. Schmidt-Trucksass, A., et al., *Arterial properties of the carotid and femoral artery in endurance-trained and paraplegic subjects.* J Appl Physiol (1985), 2000. **89**(5): p. 1956-63.

94. Rauramaa, R., et al., *Effects of aerobic physical exercise on inflammation and atherosclerosis in men: the DNASCO Study: a six-year randomized, controlled trial.* Ann Intern Med, 2004. **140**(12): p. 1007-14.
95. Ferreira, I., et al., *Longitudinal changes in .VO2max: associations with carotid IMT and arterial stiffness.* Med Sci Sports Exerc, 2003. **35**(10): p. 1670-8.
96. Howden, E.J., et al., *Cardiorespiratory fitness and cardiovascular burden in chronic kidney disease.* J Sci Med Sport, 2014.
97. Boreham, C.A., et al., *Cardiorespiratory fitness, physical activity, and arterial stiffness: the Northern Ireland Young Hearts Project.* Hypertension, 2004. **44**(5): p. 721-6.
98. Sakuragi, S., et al., *Influence of adiposity and physical activity on arterial stiffness in healthy children: the lifestyle of our kids study.* Hypertension, 2009. **53**(4): p. 611-6.
99. Melo, X., et al., *Intima-Media Thickness in 11-13 Years-Old Children: Variation Attributed to Sedentary Behavior, Physical Activity, Cardiorespiratory Fitness and Waist Circumference.* J Phys Act Health, 2014.
100. Janz, K.F., J.D. Dawson, and L.T. Mahoney, *Increases in physical fitness during childhood improve cardiovascular health during adolescence: the Muscatine Study.* Int J Sports Med, 2002. **23 Suppl 1**: p. S15-21.
101. Grontved, A., et al., *Independent and combined association of muscle strength and cardiorespiratory fitness in youth with insulin resistance and beta-cell function in young adulthood: the European Youth Heart Study.* Diabetes Care, 2013. **36**(9): p. 2575-81.
102. Grontved, A., et al., *Muscle strength in youth and cardiovascular risk in young adulthood (the European Youth Heart Study).* Br J Sports Med, 2015. **49**(2): p. 90-4.
103. Williams, M.A., et al., *Resistance exercise in individuals with and without cardiovascular disease: 2007 update: a scientific statement from the American Heart Association Council on Clinical Cardiology and Council on Nutrition, Physical Activity, and Metabolism.* Circulation, 2007. **116**(5): p. 572-84.
104. Croymans, D.M., et al., *Effects of resistance training on central blood pressure in obese young men.* J Hum Hypertens, 2014. **28**(3): p. 157-64.
105. Twisk, J.W., H.C. Kemper, and W. van Mechelen, *Tracking of activity and fitness and the relationship with cardiovascular disease risk factors.* Med Sci Sports Exerc, 2000. **32**(8): p. 1455-61.

106. WHO *Waist Circumference and Waist-Hip Ratio: Report of a WHO Expert Consultation*. 2008.
107. Kromeyer, K., et al., *Perzentile für den Body-Mass-Index für das Kindes- und Jugendalter unter Heranziehung verschiedener deutscher Stichproben.* Monatsschrift Kinderheilkunde, 2001. **149**: p. 807-818.
108. Neuhauser, H.K., et al., *Blood pressure percentiles by age and height from nonoverweight children and adolescents in Germany.* Pediatrics, 2011. **127**(4): p. e978-88.
109. Touboul, P.J., et al., *Mannheim carotid intima-media thickness and plaque consensus (2004-2006-2011). An update on behalf of the advisory board of the 3rd, 4th and 5th watching the risk symposia, at the 13th, 15th and 20th European Stroke Conferences, Mannheim, Germany, 2004, Brussels, Belgium, 2006, and Hamburg, Germany, 2011.* Cerebrovasc Dis, 2012. **34**(4): p. 290-6.
110. Oren, A., et al., *Cardiovascular risk factors and increased carotid intima-media thickness in healthy young adults: the Atherosclerosis Risk in Young Adults (ARYA) Study.* Arch Intern Med, 2003. **163**(15): p. 1787-92.
111. Jani, B. and C. Rajkumar, *Ageing and vascular ageing.* Postgrad Med J, 2006. **82**(968): p. 357-62.
112. Greenland, P., et al., *Prevention Conference V : Beyond Secondary Prevention : Identifying the High-Risk Patient for Primary Prevention : Noninvasive Tests of Atherosclerotic Burden : Writing Group III.* Circulation, 2000. **101**(1): p. e16-e22.
113. Bots, M.L., et al., *Carotid intima-media thickness measurements in intervention studies: design options, progression rates, and sample size considerations: a point of view.* Stroke, 2003. **34**(12): p. 2985-94.
114. Lamotte, C., et al., *Increased intima-media thickness of the carotid artery in childhood: a systematic review of observational studies.* Eur J Pediatr, 2011. **170**(6): p. 719-29.
115. Teynor, A., et al., *An automated, interactive analysis system for ultrasound sequences of the common carotid artery.* Ultrasound Med Biol, 2012. **38**(8): p. 1440-50.
116. Society of Atherosclerosis, I. and S. Prevention Developed in collaboration with the International Atherosclerosis, *Appropriate use criteria for carotid intima media thickness testing.* Atherosclerosis, 2011. **214**(1): p. 43-6.
117. Dawson, J.D., et al., *Risk factors associated with aortic and carotid intima-media thickness in adolescents and young*

adults: the Muscatine Offspring Study. J Am Coll Cardiol, 2009. **53**(24): p. 2273-9.
118. Lavrencic, A., et al., *Carotid intima-media thickness in young patients with familial hypercholesterolaemia.* Heart, 1996. **76**(4): p. 321-5.
119. Menees, S., et al., *Variations in carotid artery intima-media thickness during the cardiac cycle in children.* J Am Soc Echocardiogr, 2010. **23**(1): p. 58-63.
120. Mittelman, S.D., et al., *Adiposity predicts carotid intima-media thickness in healthy children and adolescents.* J Pediatr, 2010. **156**(4): p. 592-7 e2.
121. Reinehr, T., et al., *Intima media thickness in childhood obesity: relations to inflammatory marker, glucose metabolism, and blood pressure.* Metabolism, 2006. **55**(1): p. 113-8.
122. Virkola, K., et al., *Cholesterol and carotid artery wall in children and adolescents with familial hypercholesterolaemia: a controlled study by ultrasound.* Acta Paediatr, 1997. **86**(11): p. 1203-7.
123. Engelen, L., et al., *Reference intervals for common carotid intima-media thickness measured with echotracking: relation with risk factors.* Eur Heart J, 2013. **34**(30): p. 2368-80.
124. Weberruß, H., et al., *Intima-media thickness does not differ between two common carotid artery segments in children.* PLOS ONE Ultrasound Imaging, 2016.
125. Cruickshank, J.K., M. Rezailashkajani, and G. Goudot, *Arterial stiffness, fatness, and physical fitness: ready for intervention in childhood and across the life course?* Hypertension, 2009. **53**(4): p. 602-4.
126. Kocyigit, A., et al., *Relation of age and sex with carotid intima media thickness in healthy children.* Turk J Med Sci, 2014. **44**(3): p. 422-6.
127. Krebs, A., et al., *Cardiovascular risk in pediatric type 1 diabetes: sex-specific intima-media thickening verified by automatic contour identification and analyzing systems.* Pediatr Diabetes, 2012. **13**(3): p. 251-8.
128. Papadimitriou, A., *Sex differences in the secular changes in pubertal maturation.* Pediatrics, 2001. **108**(4): p. E65.
129. Pena, A.S., et al., *Lack of evidence for progression of atherosclerosis during puberty in type 1 diabetes.* Pediatr Diabetes, 2015.
130. Hidvegi, E.V., et al., *Reference values of aortic pulse wave velocity in a large healthy population aged between 3 and 18 years.* J Hypertens, 2012. **30**(12): p. 2314-21.

131. Ahimastos, A.A., et al., *Gender differences in large artery stiffness pre- and post puberty.* J Clin Endocrinol Metab, 2003. **88**(11): p. 5375-80.
132. Berenson, G.S., *Childhood risk factors predict adult risk associated with subclinical cardiovascular disease. The Bogalusa Heart Study.* Am J Cardiol, 2002. **90**(10C): p. 3L-7L.
133. Davis, P.H., et al., *Carotid Intimal-Medial Thickness Is Related to Cardiovascular Risk Factors Measured From Childhood Through Middle Age: The Muscatine Study.* Circulation, 2001. **104**(23): p. 2815-2819.
134. Li, S., et al., *Childhood cardiovascular risk factors and carotid vascular changes in adulthood: the Bogalusa Heart Study.* JAMA, 2003. **290**(17): p. 2271-6.
135. Raitakari, O.T., et al., *Cardiovascular risk factors in childhood and carotid artery intima-media thickness in adulthood: the Cardiovascular Risk in Young Finns Study.* JAMA, 2003. **290**(17): p. 2277-83.
136. Tounian, P., et al., *Presence of increased stiffness of the common carotid artery and endothelial dysfunction in severely obese children: a prospective study.* The Lancet, 2001. **358**(9291): p. 1400-1404.
137. Beauloye, V., et al., *Determinants of early atherosclerosis in obese children and adolescents.* J Clin Endocrinol Metab, 2007. **92**(8): p. 3025-32.
138. Reinehr, T., et al., *Relationship between metabolic syndrome definitions for children and adolescents and intima-media thickness.* Atherosclerosis, 2008. **199**(1): p. 193-200.
139. Aggoun, Y., et al., *Impaired endothelial and smooth muscle functions and arterial stiffness appear before puberty in obese children and are associated with elevated ambulatory blood pressure.* Eur Heart J, 2008. **29**(6): p. 792-9.
140. Di Salvo, G., et al., *Abnormal myocardial deformation properties in obese, non-hypertensive children: an ambulatory blood pressure monitoring, standard echocardiographic, and strain rate imaging study.* Eur Heart J, 2006. **27**(22): p. 2689-95.
141. Morrison, K.M., et al., *Cardiovascular risk factors and non-invasive assessment of subclinical atherosclerosis in youth.* Atherosclerosis, 2010. **208**(2): p. 501-5.
142. Freedman, D.S., et al., *The relation of obesity throughout life to carotid intima-media thickness in adulthood: the Bogalusa Heart Study.* Int J Obes Relat Metab Disord, 2004. **28**(1): p. 159-66.

143. Su, T.C., et al., *An overweight or obese status in childhood predicts subclinical atherosclerosis and prehypertension/hypertension in young adults.* J Atheroscler Thromb, 2014. **21**(11): p. 1170-82.
144. McCarthy, H.D. and M. Ashwell, *A study of central fatness using waist-to-height ratios in UK children and adolescents over two decades supports the simple message--'keep your waist circumference to less than half your height'.* Int J Obes (Lond), 2006. **30**(6): p. 988-92.
145. Ashwell, M. and S. Gibson, *A proposal for a primary screening tool: 'Keep your waist circumference to less than half your height'.* BMC Med, 2014. **12**: p. 207.
146. Cote, A.T., et al., *Obesity and arterial stiffness in children: systematic review and meta-analysis.* Arterioscler Thromb Vasc Biol, 2015. **35**(4): p. 1038-44.
147. Reneman, R.S., J.M. Meinders, and A.P. Hoeks, *Non-invasive ultrasound in arterial wall dynamics in humans: what have we learned and what remains to be solved.* Eur Heart J, 2005. **26**(10): p. 960-6.
148. Cavalcante, J.L., et al., *Aortic stiffness: current understanding and future directions.* J Am Coll Cardiol, 2011. **57**(14): p. 1511-22.
149. Laurent, S., et al., *Expert consensus document on arterial stiffness: methodological issues and clinical applications.* Eur Heart J, 2006. **27**(21): p. 2588-605.
150. Rajzer, M.W., et al., *Comparison of aortic pulse wave velocity measured by three techniques: Complior, SphygmoCor and Arteriograph.* J Hypertens, 2008. **26**(10): p. 2001-7.
151. Laurent, S., *Arterial stiffness: intermediate or surrogate endpoint for cardiovascular events?* Eur Heart J, 2005. **26**(12): p. 1152-4.
152. Kohn, J.C., M.C. Lampi, and C.A. Reinhart-King, *Age-related vascular stiffening: causes and consequences.* Front Genet, 2015. **6**: p. 112.
153. Tryggestad, J.B., et al., *Obese children have higher arterial elasticity without a difference in endothelial function: the role of body composition.* Obesity (Silver Spring), 2012. **20**(1): p. 165-71.
154. Tryggestad, J.B., et al., *Arterial compliance is increased in children with type 2 diabetes compared with normal weight peers but not obese peers.* Pediatr Diabetes, 2013. **14**(4): p. 259-66.
155. Urbina, E.M., et al., *Overweight and Hyperinsulinemia Provide Individual Contributions to Compromises in Brachial Artery*

Distensibility in Healthy Adolescents and Young Adults: Brachial Distensibility in Children. J Am Soc Hypertens, 2007. **1**(3): p. 200-207.
156. Veijalainen, A., et al., *Associations of cardiorespiratory fitness, physical activity, and adiposity with arterial stiffness in children.* Scand J Med Sci Sports, 2015.
157. Chalmers, L.J., et al., *Paradoxical increase in arterial compliance in obese pubertal children.* Angiology, 2011. **62**(7): p. 565-70.
158. Lurbe, E., et al., *Blood pressure and obesity exert independent influences on pulse wave velocity in youth.* Hypertension, 2012. **60**(2): p. 550-5.
159. Tryggestad, J.B. and K.R. Short, *Arterial compliance in obese children: implications for cardiovascular health.* Exerc Sport Sci Rev, 2014. **42**(4): p. 175-82.
160. Dangardt, F., et al., *Obese children show increased intimal wall thickness and decreased pulse wave velocity.* Clin Physiol Funct Imaging, 2008. **28**(5): p. 287-93.
161. Zieman, S.J., V. Melenovsky, and D.A. Kass, *Mechanisms, pathophysiology, and therapy of arterial stiffness.* Arterioscler Thromb Vasc Biol, 2005. **25**(5): p. 932-43.
162. Lakatta, E.G., *Arterial and Cardiac Aging: Major Shareholders in Cardiovascular Disease Enterprises: Part I: Aging Arteries: A "Set Up" for Vascular Disease.* Circulation, 2003. **107**(1): p. 139-146.
163. Safar, M.E., J. Blacher, and P. Jankowski, *Arterial stiffness, pulse pressure, and cardiovascular disease-is it possible to break the vicious circle?* Atherosclerosis, 2011. **218**(2): p. 263-71.
164. Singhal, A., et al., *Influence of leptin on arterial distensibility: a novel link between obesity and cardiovascular disease?* Circulation, 2002. **106**(15): p. 1919-24.
165. Zachariah, J.P., et al., *Circulating Adipokines and Vascular Function: Cross-Sectional Associations in a Community-Based Cohort.* Hypertension, 2016. **67**(2): p. 294-300.
166. Hvidt, K.N., et al., *Obese children and adolescents have elevated nighttime blood pressure independent of insulin resistance and arterial stiffness.* Am J Hypertens, 2014. **27**(11): p. 1408-15.
167. Cuspidi, C., et al., *Similarities and differences in structural and functional changes of left ventricle and carotid arteries in young borderline hypertensives and in athletes.* J Hypertens, 1996. **14**(6): p. 759-64.

168. Giannattasio, C., et al., *Effects of physical training of the dominant arm on ipsilateral radial artery distensibility and structure.* J Hypertens, 2001. **19**(1): p. 71-7.
169. Idris, N.S., et al., *Effect of physical activity on vascular characteristics in young children.* Eur J Prev Cardiol, 2014.
170. Meyer, A.A., et al., *Improvement of early vascular changes and cardiovascular risk factors in obese children after a six-month exercise program.* J Am Coll Cardiol, 2006. **48**(9): p. 1865-70.
171. Wunsch, R., et al., *Intima-media thickness in obese children before and after weight loss.* Pediatrics, 2006. **118**(6): p. 2334-40.
172. Li, Y., et al., *Aerobic, resistance and combined exercise training on arterial stiffness in normotensive and hypertensive adults: A review.* Eur J Sport Sci, 2015. **15**(5): p. 443-57.
173. Radtke, T., et al., *Ultra-endurance sports have no negative impact on indices of arterial stiffness.* Eur J Appl Physiol, 2014. **114**(1): p. 49-57.
174. Cornelissen, V.A., R. Buys, and N.A. Smart, *Endurance exercise beneficially affects ambulatory blood pressure: a systematic review and meta-analysis.* J Hypertens, 2013. **31**(4): p. 639-48.
175. Lee, C.D., S.N. Blair, and A.S. Jackson, *Cardiorespiratory fitness, body composition, and all-cause and cardiovascular disease mortality in men.* Am J Clin Nutr, 1999. **69**(3): p. 373-80.
176. Eisenmann, J.C., et al., *Fatness, fitness, and cardiovascular disease risk factors in children and adolescents.* Med Sci Sports Exerc, 2007. **39**(8): p. 1251-6.
177. Li, Y., et al., *Lower Body vs. Upper Body Resistance Training and Arterial Stiffness in Young Men.* Int J Sports Med, 2015. **36**(12): p. 960-7.
178. Okamoto, T., M. Masuhara, and K. Ikuta, *Upper but not lower limb resistance training increases arterial stiffness in humans.* Eur J Appl Physiol, 2009. **107**(2): p. 127-34.
179. Bernhardt, D.T., et al., *Strength training by children and adolescents.* Pediatrics, 2001. **107**(6): p. 1470-2.
180. Okamoto, T., M. Masuhara, and K. Ikuta, *Home-based resistance training improves arterial stiffness in healthy premenopausal women.* Eur J Appl Physiol, 2009. **107**(1): p. 113-7.
181. Yoshizawa, M., et al., *Effect of 12 weeks of moderate-intensity resistance training on arterial stiffness: a randomised*

controlled trial in women aged 32-59 years. Br J Sports Med, 2009. **43**(8): p. 615-8.
182. Cortez-Cooper, M.Y., et al., *The effects of strength training on central arterial compliance in middle-aged and older adults.* Eur J Cardiovasc Prev Rehabil, 2008. **15**(2): p. 149-55.
183. Fahs, C.A., et al., *Muscular strength is inversely associated with aortic stiffness in young men.* Med Sci Sports Exerc, 2010. **42**(9): p. 1619-24.
184. Neuhauser, H. and M. Thamm, *[Blood pressure measurement in the German Health Interview and Examination Survey for Children and Adolescents (KiGGS). Methodology and initial results].* Bundesgesundheitsblatt Gesundheitsforschung Gesundheitsschutz, 2007. **50**(5-6): p. 728-35.
185. den Ruijter, H.M., et al., *Common carotid intima-media thickness does not add to Framingham risk score in individuals with diabetes mellitus: the USE-IMT initiative.* Diabetologia, 2013. **56**(7): p. 1494-502.
186. Bots, M.L., et al., *Common Carotid Intima-Media Thickness Measurements Do Not Improve Cardiovascular Risk Prediction in Individuals With Elevated Blood Pressure: The USE-IMT Collaboration.* Hypertension, 2014.
187. Spence, J.D., *Measurement of intima-media thickness vs. carotid plaque: uses in patient care, genetic research and evaluation of new therapies.* Int J Stroke, 2006. **1**(4): p. 216-21.
188. Katzmarzyk, P.T., et al., *Physical Activity, Sedentary Time, and Obesity in an International Sample of Children.* Med Sci Sports Exerc, 2015. **47**(10): p. 2062-9.
189. Freedman, D.S., et al., *The relation of childhood BMI to adult adiposity: the Bogalusa Heart Study.* Pediatrics, 2005. **115**(1): p. 22-7.
190. Hasselstrom, H., et al., *Physical fitness and physical activity during adolescence as predictors of cardiovascular disease risk in young adulthood. Danish Youth and Sports Study. An eight-year follow-up study.* Int J Sports Med, 2002. **23 Suppl 1**: p. S27-31.

8 APPENDIX

8.1 Publication 1

Weberruß H, Pirzer R, Böhm B, Elmenhorst J, Dalla Pozza R, Netz H, Oberhoffer, R. Increased intima-media thickness is not associated with stiffer arteries in children. Atherosclerosis. 2015;242(1): 48-55. doi: 10.1016/j.atherosclerosis

8.2 Publication 2

Weberruß, H., Pirzer, R., Böhm, B., Dalla Pozza, R., Netz, H., & Oberhoffer, R. (2016). Intima-media thickness and arterial function in obese and non-obese children. BMC Obesity, 3(1). doi:10.1186/s40608-016-0081-9

8.3 Publication 3

Weberruß, H., Pirzer, R., Schulz, T., Böhm, B., Dalla Pozza, R., Netz, H., & Oberhoffer, R. (2016). Reduced arterial stiffness in very fit boys and girls. Cardiology in the Young. 2016 Mar 29:1-8. doi: 10.17/S1047951116000226