Original Articles

Association of inflammation with adiposity related metabolic derangements and waist-to-height ratio in predicting insulin resistance and low-grade inflammation among 8-9-year-old children in an urban area of Sri Lanka

***Kalaichelvi Thillan[1], Pulani Lanerolle[1], Dulani Samaranayake[2], Tharanga Thoradeniya[1], Pujitha Wickramasinghe[3]**

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Abstract

Background: The inflammatory link between high adiposity and metabolic derangements and the usefulness of waist-to-height ratio (WHtR) in screening low grade inflammation and insulin resistance (IR) have not been documented among Sri Lankan children.

Objectives: To describe association of inflammation with adiposity related metabolic derangements and develop cut-off values for WHtR in predicting IR and low-grade inflammation among 8-9-year-old children from an urban area of Sri Lanka

Method: This paper analysed data collected for a case-control study where children with high body fat (BF) (n=147; male-76) and normal BF (n=152; male-75) were recruited from primary schools in the Colombo Municipal Council area. Anthropometry, % BF, fasting blood sugar, insulin, total cholesterol, high density lipoprotein-cholesterol, triglyceride, low density lipoprotein-cholesterol, high sensitivity C-reactive protein (hs-CRP), leptin, adiponectin and homeostasis model of IR were analysed. In addition, cut-offs of WHtR were developed to predict IR (>1.7) and low-grade inflammation (hs-CRP>1mg/L).

Results: High BF group had significantly higher metabolic and inflammatory parameters than normal BF group in both sexes except adiponectin which was low in high BF group. Metabolic parameters were significantly higher among those with IR in males and females. Children with inflammation had higher %BF, WHtR and metabolic parameters than those with no inflammation. WHtR of 0.47 and 0.45 to detect inflammation, and 0.48 and 0.46 to detect IR, among males and females respectively showed good sensitivity and specificity.

Conclusions: Inflammation and metabolic derangements increase with BF. WHtR is useful in screening for development of inflammation and IR.

(Key words: Inflammation, Metabolic derangements, IR, WHtR)

Introduction

Metabolic consequences associated with obesity begin in childhood and predispose to cardiovascular disease in adult life[1]. Clustering of these metabolic derangements is termed metabolic syndrome (MetS). In 2007, International Diabetic Federation outlined criteria to diagnose MetS in 10-16-year-old children but suggested not to diagnose MetS in children under 10 years but only to identify the metabolic derangements[2]. Expansion of adipose tissue initiates cascades of inflammatory events resulting in defective insulin signalling pathways leading to insulin resistance (IR)[3]. Impairment in insulin action due to resistance results in metabolic abnormalities[4]. Obesity-associated metabolic derangements[5] and MetS[6] have been reported among Sri Lankan children. Further, a higher IR has been reported among Sri Lankan children with higher body mass index (BMI) and low birth weight (LBW)[7]. However, the inflammatory link between obesity and metabolic derangements related to body fat has not been documented in Sri Lankan children and they have been reported to have higher fat mass from a young age[8]. Biochemical analysis of inflammatory markers and insulin is costly and time-consuming. Therefore, availability of a simple anthropometric tool for screening would be valuable. BMI, a simple widely used index to assess

[1]*Department of Biochemistry and Molecular Biology, Faculty of Medicine, University of Colombo, Sri Lanka,* [2]*Department of Community Medicine, Faculty of Medicine, University of Colombo, Sri Lanka,* [3]*Department of Paediatrics, Faculty of Medicine, University of Colombo, Sri Lanka*
**Correspondence:* chelvi.thillan@yahoo.co.in
https://orcid.org/0000-0002-3699-0160
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nutritional status, does not differentiate fat and fat-free mass. WHtR is a simple, cost-effective, non-invasive anthropometric index of measuring abdominal obesity. WHtR cut-offs of 0.51 and 0.49 have been reported to detect MetS for boys and girls aged 5-15-years[9]. However, no studies have assessed the effectiveness of WHtR in predicting IR and inflammation in Sri Lankan children.

Objectives

To describe the association of inflammation with adiposity related metabolic derangements and to develop cut-off values for WHtR in predicting IR and low-grade inflammation among 8-9-year-old children from an urban area of Sri Lanka

Method

Sample size: This paper presents data from a case-control study conducted from August 2015 to November 2016 involving 8-9-year-old primary school children within Colombo Municipal Council (CMC) area to explore the association between adiposity and micronutrient status. Therefore, sample size was based on available local and international data on micronutrient deficiencies[10-14] among obese and normal-weight children. Highest sample size estimated per group was 81 (α error 0.05; 80% statistical power), considering a 10% dropout rate. Four groups were formed stratified by sex in both cases and controls. Further, sample size adequacy was checked for analysis of inflammatory and metabolic derangement factors (α error 0.05; power 80%) using published mean values of relevant parameters among normal weight and overweight/obese children[15-16].

Participant recruitment: Thirty-seven schools were randomly selected from 110 schools within CMC area. Overweight/obese and normal weight children were screened based on the BMI (overweight >+1SD and < +2SD and obese ≥+2SD for each sex, based on BMI for age WHO 2007 standards) during the routine school medical inspection. Recruitment was done with prior approval obtained from the Ministry of Education and school principals. Selected children and their parents were invited to the Clinical Laboratory of the Professorial Paediatric Unit at the Lady Ridgeway Hospital for Children, Colombo and recruited following informed written consent. BF, anthropometry measures and blood samples were collected. Children with high and normal BF were categorized based on the percentage BF cut offs associated with MetS[17]. Children with BF higher than the cut-off value (%BF >28.6% in males and >33.7% in females) were defined as cases (n=147; male 76) and the children with BF below the cut-off value (% BF ≤28.6% in males and ≤33.7% in females) were defined as controls (n=152; males 75).

Inclusion and exclusion criteria: Children aged 8-9 years who were residents of CMC area and identified to have normal and high % BF by the cut off values published for Sri Lankan children[17] were included. Children with chronic illness, birth defects, restricted diet and infection/inflammation within preceding two weeks of study were excluded.

Anthropometric and BF measurements: Height was measured to the nearest 0.1cm without shoes using a portable stadiometer (SECA 225; Germany); weight was measured using an electronic weighing scale (SECA 803; Germany) to the nearest 0.1kg with light clothes. BMI was calculated as weight (kg) divided by height squared (m). Waist circumference (WC) was measured horizontally at the level of the midpoint between the lower border of costal margin and highest point of the iliac crest in the mid axillary line, using a non-stretchable tape to the nearest 0.1cm at the end of expiration. WHtR was calculated as waist circumference (cm) divided by height (cm). BF was measured by bioelectrical impedance analysis (BIA) technique using an eight electrode multi-frequency InBody 230 analyzer (Biospace Co., Ltd.; Seoul, Korea). Children with minimum clothes without any metal accessories (belts, bangles, wrist watches) stood on the platform barefoot, placing the feet on the electrodes. The handheld electrodes were placed maximally away from the body after properly placing the thumb and the other four fingers on the respective electrodes. Measurements were taken within three minutes while the child was standing still. BF measurements and other required parameters were recorded.

Blood sample collection: A blood sample (8ml) following an overnight fast (12 hours) was drawn by an experienced nursing officer under sterile conditions.

Biochemical analysis: The total cholesterol (TC), triglyceride (TG), and high-density lipoprotein-c (HDL-c) were analysed by Dimension Clinical Biochemistry system (Dimension X^{pand}Plus Random access automated clinical chemistry analyzer, Siemens Healthcare Diagnostics Inc, USA) with ready to use reagent pack. Friedwald formula [(TC-HDL)-TG/5] was used to calculate LDL-c. Non-HDL cholesterol was calculated by subtracting HDL-c from TC. Cut off value for serum lipid levels were defined as TC ≥200mg/dL, TG ≥100mg/dL, HDL ≤45mg/dL, non-HDL-c ≥144mg/dL and LDL-c ≥130mg/dL)[18]. Fasting blood sugar (FBS) was analysed by Dimension Clinical Biochemistry system (Dimension X^{pand} Plus Random access automated Clinical chemistry analyzer) using hexokinase method. FBS was defined as ≥100mg/dL[19]. Serum insulin was analysed by automated random access IMMULITE® 1000 immunoassay system (Siemens Healthcare Diagnostics Inc. USA) using solid phase enzyme labeled two site sandwich chemiluminescent immunometric assay. IR was assessed using Homeostasis Model Assessment of Insulin Resistance (HOMA-IR)[20]. HOMA IR was

calculated as fasting blood sugar (mmol/L) × fasting insulin (microU/ml) ÷ 22.5 and the elevated level of HOMA IR was defined as >1.7 for pre pubertal children for both sexes[21]. hs-CRP was assessed by single step immunometric assay (Siemens Healthcare Diagnostics Inc, USA) while leptin and adiponectin were assessed by enzyme-linked immunosorbent assay (ELISA) (EIA-2395, DRG International, Inc, USA), EIA-4574, DRG International, Inc, USA). Adiponectin analysis was performed in a sub sample of children (n=137) and the leptin: adiponectin ratio (leptin/adiponectin) was calculated. The elevated levels of hs-CRP and leptin were defined as >1mg/dL[22] and 13.4ng/mL[23] respectively. Due to the unavailability of well-established cut off values, the reference values of adiponectin (2.2µg/mL)[24] and leptin: adiponectin ratio (males-1.28 ng/µg, females-0.87 ng/µg[25] were used to categorize groups.

Ethical issues: Study was approved by the Ethics Review Committees of Faculty of Medicine, University of Colombo (EC-14-168), and LRHC, Colombo. Informed written consent was obtained from parents of participating children.

Statistical analysis: This was done using SPSS version 20. Analysis was separately done for males and females. Continuous variables were compared using independent sample t-test and non-parametric Mann–Whitney U test. Categorical variables were compared using Chi-square test between groups. Metabolic derangements were compared across normal and high categories of both IR and hs-CRP among both sexes. Receiver operating characteristic (ROC) curves were drawn to determine cut-offs for WHtR to predict IR (>1.7) and inflammation (hs-CRP>1mg/L). Optimum cut-off points were separately determined for males and females. Subjects with CRP>10mg/L were excluded in the analysis of hs-CRP to avoid possibility of having concurrent acute inflammatory conditions such as infections. Statistical significance was set at $p<0.05$.

Results

Data of 299 children were used in final analysis. Comparison of anthropometry, body composition, metabolic and inflammatory parameters of high and normal BF groups according to sex are given in Table 1.

Table 1: ***Comparison of anthropometry, body composition, metabolic and inflammatory parameters in high and normal body fat groups according to sex***

Characteristic	Gender	Children with high %BF Males n=76 (% BF>28.6) Females n=71 (%BF>33.7)	Children with normal %BF Males n=75 (≤28.6%) Females n=77 (≤33.6%)
Age (years) - Mean ± SD	Male	9.1 ± 0.32	9.2 ± 0.36
	Female	9.1 ± 0.27	9.1 ± 0.29
Anthropometry and body composition			
Height (metres) - Mean ± SD	Male	1.4 ± 0.06	1.31 ± 0.07#
	Female	1.3 ± 0.05	1.30 ± 0.06#
Weight (kg) - Mean ± SD	Male	39.50 ± 1.18	26.10 ±1.22#
	Female	37.76 ± 1.16	25.50 ± 1.18#
Body mass index (kg/m^2) – Median (IQR)	Male	21.03 (19.2, 23.2)	14.63 (13.8, 17.1) #
	Female	20.66 (19.3, 22.0)	14.96 (13.7, 16.4) #
Waist circumference (cm) - Mean ± SD	Male	71.09 ± 1.08	55.37 ± 1.11#
	Female	72.59 ± 1.12	54.82 ± 1.12#
Waist- to-height ratio - Mean ± SD	Male	0.53 ± 1.11	0.42 ± 1.09#
	Female	0.53± 1.08	0.43± 1.09#
Percentage of body fat – Median (IQR)	Male	35.92 (31.7, 40.2)	16.13 (12.8, 23.6) #
	Female	38.0 (35.4, 41.2)	20.26 (15.3, 27.0) #
Metabolic risk factors			
Fasting blood sugar (mg/dL) - Mean ± SD	Male	93.50 ± 1.12	90.99 ± 1.08
	Female	92.47 ± 1.13	88.92 ± 1.10*
Fasting insulin (µLU/mL) – Median (IQR)	Male	5.88 (3.6, 11.6)	2.09 (1.8, 3.9) #
	Female	7.98 (5.3, 12.8)	3.00 (2.0, 5.1) **
HOMA IR[20] – Median (IQR)	Male	1.36 (0.8, 2.7)	0.48 (0.4,0.9) #
	Female	1.83 (1.2, 2.7)	0.62 (0.4, 1.1) #
Total cholesterol (mg/dL) – Median (IQR)	Male	184 (168, 198)	174 (159, 193)
	Female	185 (168, 206)	183 (159, 202)
High density lipoprotein cholesterol (mg/dL) – Median (IQR)	Male	45 (40, 52)	51 (42.5, 59.5) **
	Female	42 (37, 49)	49 (42, 56) #
Triglyceride (mg/dL) - Mean ± SD	Male	80.37 ± 1.60	61.11 ± 1.49#
	Female	87.80 ± 1.56	69.33 ± 1.51#
Low density lipoprotein cholesterol (mg/dL) – Median (IQR)	Male	120 (106.2, 132)	114 (97, 122.8)
	Female	122 (108, 139.6)	114.8 (99. 4,137)
Non - high density lipoprotein cholesterol (mg/dL) - Mean ± SD	Male	133.96 ± 1.50	124.77 ± 1.29*
	Female	144.91 ± 1.33	129.91 ± 1.59**
Inflammatory markers			
High sensitivity C-reactive protein (mg/L)[1] – Median (IQR)	Male	1.34 (0.4, 3.3)	0.15 (0.06, 0.4) #
	Female	1.22 (0.5, 2.4)	0.17 (0.05,1.02) #
Leptin (ng/mL) – Median (IQR)	Male	9.87 (5.5, 14.8)	2.18 (1.0, 5.3) #
	Female	15.10 (9.7, 21.9)	3.66 (1.9, 6.6) #
Adiponectin (µg/mL) – Median (IQR)	Male	2.87 (1.6, 3.7)	5.06 (3.1, 7.7) **
	Female	3.14 (2.3, 4.6)	5.28 (3.2, 7.3) **
Leptin: Adiponectin (ng/µg) – Median (IQR)	Male	4.39 (2.4, 10.0)	0.46 (0.2, 1.8) #
	Female	5.41 (2.4, 11.2)	0.63 (0.4, 1.9) #

*HOMA IR – Homeostasis model assessment of insulin resistance, [1]hs-CRP >10mg/L excluded in analysis of hs-CRP, Independent sample t-test and Mann -Whitney U test with median (inter quartile range) were used in comparison of parameters between BF groups within each sex. Statistically significant - p<0.001#, p<0.01**, p<0.05**.*

Age differences were not seen between groups of both sexes. As expected, children with high BF had higher anthropometric and body composition measurements. The high BF groups of both sexes had significantly higher metabolic derangement factors including fasting insulin, HOMA IR, HDL-c, TG and non-HDL-c compared to controls except TC and LDL-c. Similarly, the high BF group had significantly higher levels ($p<0.001$) of hs-CRP (>1mg/L) and leptin compared to normal BF group in both sexes. Adiponectin levels were significantly low among children with high BF in both sexes compared to normal BF group ($p<0.01$). In contrast, high BF group had a significantly higher leptin/adiponectin ratio ($p<0.001$) (Table 1).

The prevalence of metabolic derangements measured by HDL (≤45mg/dL), TG (≥100mg/dL), and HOMA IR (>1.7) was significantly high among males and females with high BF, while a significantly higher prevalence of non-HDL-c (≥144mg/dL) and LDL-c (≥130mg/dL) was seen among males and FBS (≥100mg/dL) in females with high BF compared to the normal fat group. Similarly, the prevalence of inflammation measured by hs-CRP (>1mg/L) and leptin (>13.4ng/mL) was higher among the children with high BF compared to the normal group. However, a higher percentage of males ($p>0.05$) and females ($p<0.05$) with high BF had low levels of adiponectin. In contrast, a higher proportion of high fat children in both sexes had a higher leptin/adiponectin ratio compared to their leaner counterparts ($p<0.001$) (Table 2).

Table 2 ***Comparison of metabolic and inflammatory markers in high and normal body fat groups***

Marker	Male			Female		
	High %BF (>28.6) (n=76)	**Normal %BF (≤28.6) (n=75)**	**p**	**High %BF (>28.6) (n=76)**	**Normal %BF (≤28.6) (n=75)**	**p**
Total cholesterol (mg/dL)						
<200 – n (%)	60 (80.0)	59 (80.8)	0.9	52 (73.2)	56 (74.7)	0.844
≥200 – n (%)	15 (20.0)	14 (19.2)		19 (26.8)	19 (25.3)	
HDL-c (mg/dL) – n (%)						
≤45 – n (%)	40 (53.3)	23 (31.5)	**0.007**	47 (66.2)	25 (33.3)	**<0.001**
>45 – n (%)	35 (46.7)	50 (68.5)		24 (33.8)	50 (66.7)	
Non-HDL-c (mg/dL)						
<144 – n (%)	45 (60.0)	60 (82.2)	**0.003**	37 (52.1)	48 (64.0)	0.145
≥144 – n (%)	30 (40.0)	13 (17.8)		34 (47.9)	27 (36.0)	
Triglycerides (mg/dL						
<100 – n (%)	52 (69.3)	66 (90.4)	**0.001**	43 (60.6)	61 (81.3)	**0.006**
≥100 – n (%)	25 (30.7)	07 (09.6)		28 (39.4)	14 (18.7)	
LDL-c (mg/dL)						
<130 – n (%)	50 (66.7)	63 (86.3)	**0.005**	45 (63.4)	52 (69.3)	0.446
≥130 – n (%)	25 (33.3)	10 (13.7)		26 (36.6)	23 (30.7)	
Fasting blood sugar						
<100mg/dL – n (%)	56 (73.7)	63 (85.1)	0.083	55 (77.5)	69 (89.6)	**0.045**
≥100mg/dL – n (%)	20 (26.3)	11 (14.9		16 (22.5)	08 (10.4)	
HOMA IR[20]						
≤1.7 – n (%)	45 (60.0)	72 (97.3)	**<0.001**[b]	32 (47.1)	69 (92.0)	**<0.001**
>1.7 – n (%)	30 (40.0)	02 (02.7)		36 (52.9)	06 (08.0)	
[1]hs-CRP						
≤1mg/L – n (%)	34 (44.2)	64 (82.1)	**<0.001**	31 (40.8)	64 (76.2)	**<0.001**
>1mg/L – n (%)	43 (55.8)	14 (17.9)		45 (59.2)	20 (23.8)	
Leptin (Lep)						
≤13.4ng/mL – n (%)	57 (71.3)	71 (88.8)	**0.006**	32 (40.5)	74 (88.1)	**<0.001**
>13.4ng/mL – n (%)	23 (28.8)	09 (11.3)		47 (59.5)	10 (11.9)	
Adiponectin (Adi)						
2.2 µg/mL	23 (65.7)	29 (85.3)	0.059	25 (80.6)	33 (97.1)	**0.033**[b]
<2.2µg/mL	12 (34.3)	05 (14.7)		06 (19.4)	01 (02.9)	
[24]Lep/Adi ratio (ng/µg)						
Normal	06 (17.1)	24 (70.6)	<**0.001**	0 (0.0)	22 (64.7)	**<0.001**[b]
High	29 (82.9)	10 (29.4)		34 (100.0)	12 (35.3)	

HOMA IR: Homeostasis model assessment of insulin resistance; hs-CRP: High sensitivity C-reactive protein. [1]hs-CRP >10mg/L excluded in the analysis of hs-CRP; Pearson Chi-square value -statistically significant at p<0.05; b- Fisher's Exact Test; Leptin/adiponectin ratio was categorized as male-1.28 ng/µg, female-0.87 ng/µg; Adiponectin measurements were performed only in a sub sample. Male- (High BF-35, Normal BF-34) Female- (High BF-34, Normal BF-34)

Table 3 shows the comparison of metabolic risk factors and hs-CRP across the normal and high IR categories among males and females. The children with high IR (>1.7) had significantly higher median levels of metabolic parameters and hs-CRP than those with normal IR among both sexes.

Table 3: ***Comparison of distribution of metabolic risk factors in high and normal insulin resistance groups according to sex***

Metabolic risk factor	Males			Females		
	Normal HOMA (IR≤1.7) n=116 - Median (IQR)	**High HOMA (IR>1.7) n=32 - Median (IQR)**	**p**	**Normal HOMA (IR≤1.7) n=100 -Median (IQR)**	**High HOMA (IR>1.7) n=42 -Median (IQR)**	**p**
Fasting blood sugar (mg/dL)	90 (85.5, 98.0)	96.5 (90.5, 105.75)	**<0.001**	87.0 (82, 94)	92.5 (86.75, 105.50)	**<0.001**
Total cholesterol (mg/dL)	176.5 (159.5, 192.75)	193.5 (175.0, 202.0)	**0.007**	181.5 (162, 197.75)	185.5 (173, 215.25)	**0.022**
HDL-c (mg/dL)	48.5 (41.0, 56.0)	44 (36.75, 50.75)	**0.006**	48 (41, 54)	41 (36. 46)	**0.001**
Non-HDL-c (mg/dL)	128 (108.0, 142.75)	144 (129.25, 157.0)	**<0.001**	133 (115, 151)	149 (133, 173.5)	**0.001**
Triglyceride (mg/dL)	63 (48, 83)	96 (71, 148.25)	**<0.001**	73.5 (53, 91.75)	91 (71.25, 133.25)	**<0.001**
LDL-C	114.6 (97.7, 127.15)	123.3 (112.4, 136.05	**0.008**	116.7 (101.6, 135)	125.5 (108.55, 147.95	**0.045**
hs-CRP	0.24 (0.08, 1.14)	1.88 (0.66, 5.22)	**<0.001**	0.34 (0.08, 1.37)	1.23 (0.46, 2.74)	**<0.001**

HOMA IR: Homeostasis model assessment of insulin resistance, HDL-c: High density lipoprotein cholesterol, LDL-c: Low density lipoprotein cholesterol. hs-CRP (mg/L): High sensitivity C-reactive protein, IQR: Inter-quartile range

Further, the children with elevated levels of hs-CRP (>1mg/L) had higher % BF, WHtR (p<0.001) and metabolic derangements among males [HDL-c (p<0.01), non-HDL-c (p<0.01), TG (p<0.01), LDL-c (p<0.01), HOMA IR (p<0.001)] and females [TG (p<0.05), HOMA IR (p<0.01)] compared to the children with normal hs-CRP (≤1mg/L) (Table-4).

Table 4: ***Comparison of body composition, anthropometric and metabolic risk parameters in non-inflammatory and inflammatory groups among males and females***

Characteristic	Males			Females		
	Non-inflammatory group hs-CRP≤1mg/L (n=93) Median (IQR)	Inflammatory group hs-CRP>1mg/L (n=52) Median (IQR)	p	Non-inflammatory group hs-CRP≤1mg/L (n=86) Median (IQR)	Inflammatory group hs-CRP>1mg/L (n=57) Median (IQR)	p
Percentage body fat	22.72 (14.36, 30.71)	36.20 (28.92, 41.18)	**<0.001**	25.32 (15.63, 35.14)	36.0 (29.85, 40.45)	**<0.001**
Waist- to-height ratio	0.44 (0.40, 0.49)	0.53 (0.47, 0.57)	**<0.001**	0.44 (0.41, 0.49)	0.51 (0.47, 0.54)	**<0.001**
Fasting blood sugar (mg/dL)	91 (86, 98)	91.5 (87, 100.5)	0.420	90 (84, 94.75)	92 (85. 98)	0.295
Total cholesterol (mg/dL)	173 (159.5,193)	183 (162, 198)	0.144	182.5 (161.5, 204.5)	185 (165.5, 199.5)	0.490
HDL-c (mg/dL)	51 (43, 56)	43 (39, 52)	**0.003**	47 (39, 54)	45 (37.5, 50.0)	0.060
Non-HDL-c (mg/dL)	125 (107, 145)	139 (119, 154)	**0.006**	134 (114.75, 155.25)	142 (123, 152.5)	0.110
Triglyceride (mg/dL)	61 (46.25,81.75)	81 (54, 106)	**0.002**	72.5 (54.5, 88.75)	89 (63.0, 110.50)	**0.011**
LDL-c (mg/dL)	113.10 (97.7, 126.5)	120 (103, 133)	**0.048**	114.8 (100.6, 137.55)	124.40 (108.8, 142)	0.155
[20]HOMA IR	0.73 (0.43, 1.19)	1.29 (0.58, 2.69)	**<0.001**	0.89 (0.49, 1.45)	1.40 (0.69, 2.51)	**0.002**

HDL-c: High density lipoprotein cholesterol, LDL-c: Low density lipoprotein cholesterol, HOMA IR: Homeostatic model of insulin resistance, hs-CRP (mg/L): high sensitivity C reactive protein. hs-CRP >10mg/L excluded in the analysis of hs-CRP

The correlation analysis showed a significant correlation of IR (HOMA) and hs-CRP levels with WHtR among males and females (Figure 1).

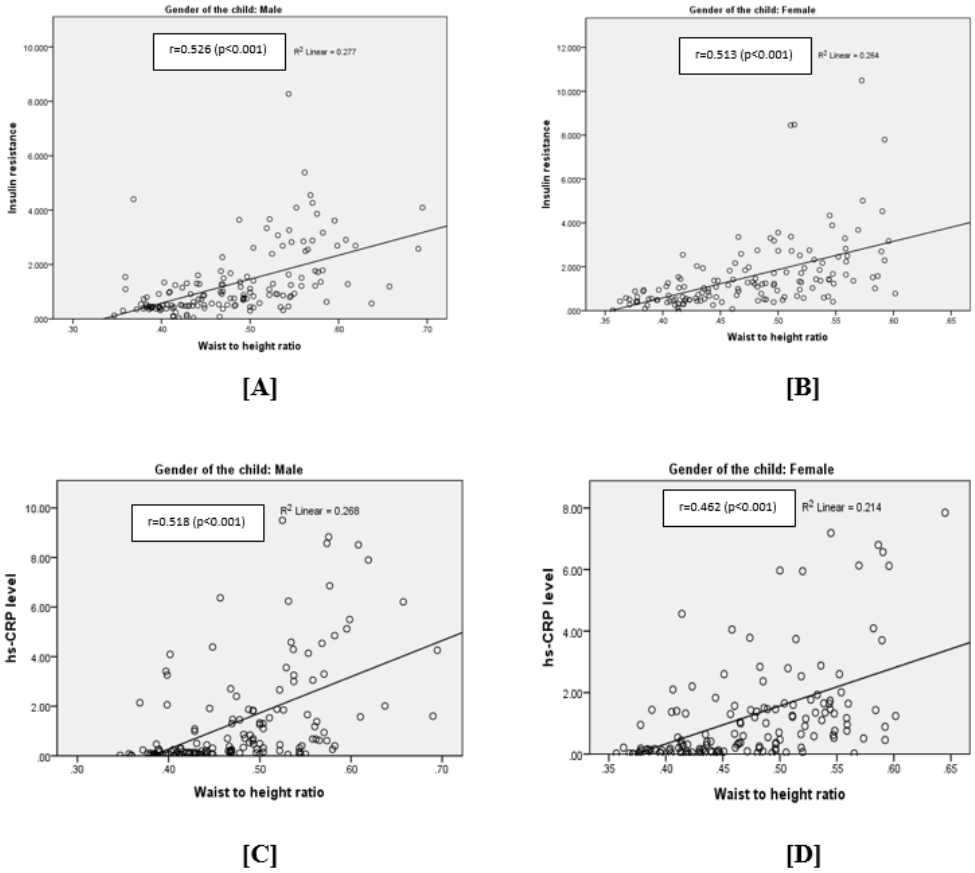

Figure 1: ***Correlation plots of waist-to-height ratio against insulin resistance (A, B) and waist-to-height ratio against high sensitivity C-reactive protein (C, D) among males and females***

Based on the ROC analysis, the optimum cut-off values of WHtR in predicting IR (> 1.7) for males and females were 0.4829 (sensitivity: 90.6%, specificity: 65.8%) and 0.4618 (sensitivity: 90.5%, specificity: 60.0%) respectively (Figure 2, Table 5). Similarly, the cut off values of WHtR in predicting inflammation (hs-CRP>1mg/L) were 0.4679 (sensitivity: 82.7%, specificity: 63.4%) and 0.4510 (sensitivity: 84.2%, specificity: 58.0%) (Figure 2, Table 5).

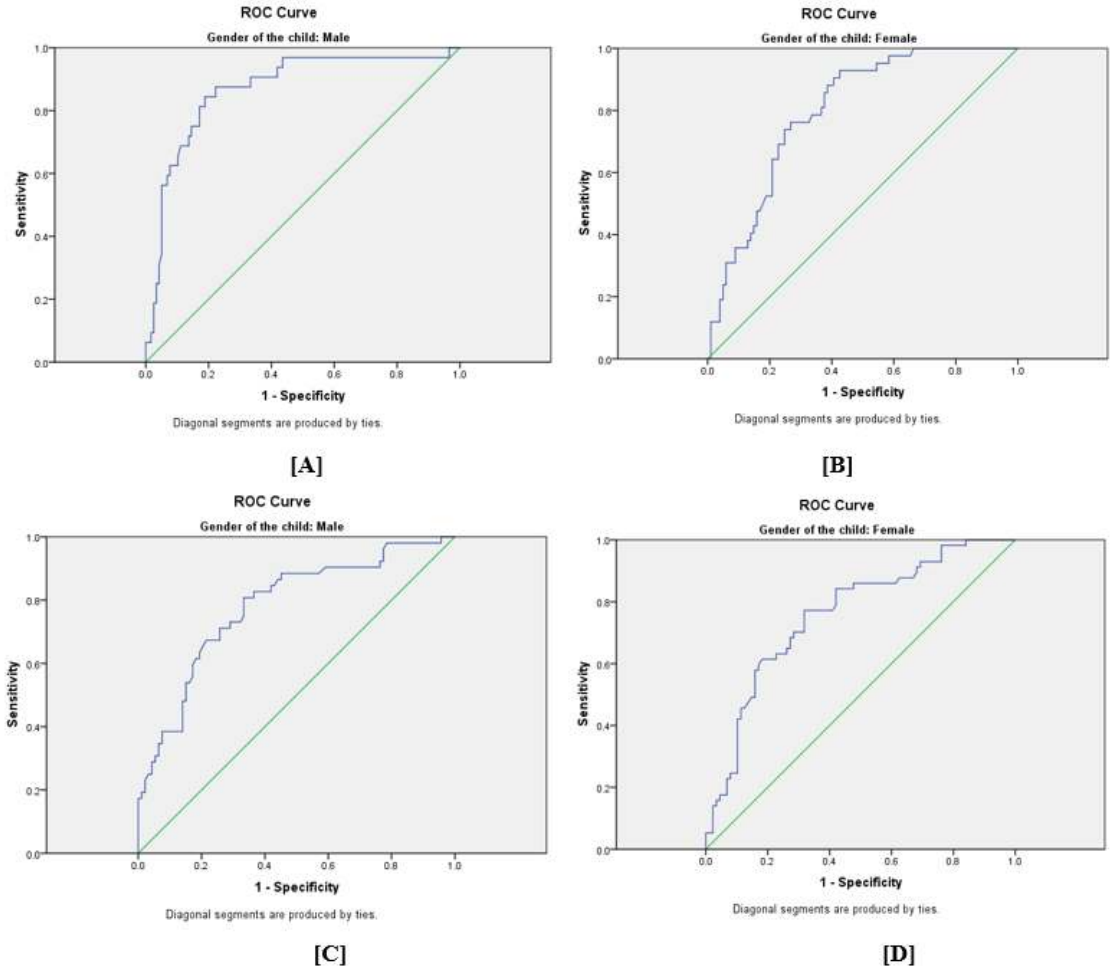

Figure 2: Receiver operating characteristic curves for the cut-off value of waist-to-height ratio in predicting insulin resistance among males (A) and females (B) and inflammation among males (C) and females (D)

Table 5: ***The determined cut-off points of waist-to-height ratio (WHtR) and related measure in predicting insulin resistance (IR) >1.7 inflammation (high sensitivity C-reactive protein >1mg/L).***

	Cut off (WHtR)	AUC	Standard error	95% CI	p-value	Sensitivity %	Specificity %
Insulin resistance (IR)							
Male	0.4829	0.863	0.042	0.795-0.942	<0.001	90.6	65.8
Female	0.4618	0.799	0.037	0.727-0.872	<0.001	90.5	60.0
Inflammation							
Male	0.4679	0.781	0.040	0.702-0.860	<0.001	82.7	63.4
Female	0.4510	0.760	0.041	0.680-0.840	<0.001	84.2	58.0

AUC: Area under curve, CI: Confidence interval. IR measured as HOMA IR[20]: Homeostasis model assessment of insulin resistance, hs-CRP: High sensitivity C-reactive protein hs-CRP >10mg/L excluded in the analysis of hs-CRP.

Discussion

The present study describes the role of inflammation as a link between high adiposity and metabolic risk among 8-9-year-old Sri Lankan children. It furthers the understanding of metabolic risk in young children. In the present study, a high prevalence of metabolic risk was witnessed among the children with high BF compared to those with normal BF in both sexes. Similarly, an increasing trend of cardiovascular risk with an unfavourable adiposity profile has been reported among pre-pubertal children[26]. However, a significantly higher prevalence of non-HDL-c and LDL-c was only observed among males and FBS in females with high BF than normal BF children. Further, the influence of sex hormone-binding globulin on adiposity and lipid profile has also been documented among pre-pubertal children[27]. The present study demonstrated higher metabolic risk factors among high BF groups compared to the normal BF groups in both sexes except for LDL-c, TC and FBS in males. A significantly high HDL-c levels of the

normal BF group may have contributed to the TC, which could be the reason for not observing a clear difference in TC between groups.

Unlike in adults, IR is still under investigation in the paediatric population. IR associated metabolic derangements have been documented in pre-pubertal children[28]. The increased metabolic derangements in children with high BF seen in the present study could be due to the higher IR. Further, the higher metabolic derangements seen in children with high IR indicates the underlying IR associated with metabolic derangements in this study population. IR is known to lead to the impairment of the regulatory role of insulin on lipoprotein metabolism and lipoprotein lipase, which involves the hydrolysation of lipoproteins[29]. Family history for obesity was not assessed in this study and may have influenced the relationship with metabolic risk[30].

Similar to metabolic derangements, a higher low grade inflammatory state was also observed among children with high BF than children with normal BF. The increased inflammatory parameters and low adiponectin (anti-inflammatory marker) in children with high BF indicate the underlying inflammation associated with high BF. Leptin is a pro-inflammatory adipokine that increases in proportion to the fat mass[31]. The higher leptin: adiponectin ratio indicates the leptin adiponectin imbalance[25].

Further, the higher levels of IR, % BF and WHtR in the children with elevated hs-CRP (>1mg/L) than the children with normal hs-CRP (≤1 mg/L) confirm the inflammatory link between high adiposity and metabolic derangements seen in this study population. The role of inflammation in the development of IR[32] and the link between IR and metabolic risk factors[33] have been documented in previous studies. Similarly, in the present study, the increased inflammatory and metabolic markers associated with IR as described above may indicate that the high adiposity induced inflammation leads to IR, which could be the underlying cause for the development of metabolic derangements observed among the children of this study population. Therefore, the inflammation and IR would be the initial steps in the development of obesity related metabolic derangements that could be used to detect the onset of obesity-related metabolic derangements. However, measurements of these parameters are lab oriented and costly.

WHtR as a simple anthropometric measure is associated with inflammation and IR. It is a cost-effective, simple and straightforward measure compared to biochemical analysis. Further, WHtR has been suggested as a useful tool to provide screening for IR in children and adolescents[34]. However, no studies have assessed the effectiveness of WHtR in predicting IR and inflammation in Sri Lankan children. While cut-offs of IR for the paediatric population are currently under debate, the cut-off we have chosen to define IR (1.7) has been reported to have the best threshold (sensitivity-86, specificity-67%) to diagnosis MetS among pre-pubertal children[21]. Further, hs-CRP has been reported as a better predictor of obesity among children[35] and an increased risk of hs-CRP concentration (>1.0mg/L) (hazard ratio 2.29) has been reported in obese children from the age of 3 years through the adolescent period[22]. Hence, in the present study, we have used hs-CRP>1mg/L as a cut off to define inflammation associated with high adiposity.

In the present study, we have identified 0.47 (sensitivity-82.7% specificity-63.7%) and 0.45 (sensitivity-84.2%, specificity-58%) as optimum cut-off values of WHtR for males and females respectively to predict inflammation. Similarly, 0.48 (sensitivity-90.6% specificity-65.8%) and 0.46 (sensitivity-90.5% specificity-60%) were identified as optimum cut-off values in predicting IR in males and females respectively in this study population. The higher sensitivity indicates the screening ability of the developed cut-offs. Further, the WHtR cut-off points of 0.51 (males) and 0.49 (females) have been reported to predict MetS in Sri Lankan children in the range of 5 to 15 years[9]. This gradual increase in the cut off values of WHtR in predicting inflammation, IR and MetS further highlight the sequence of events that occur with the gradual increase in abdominal adiposity with time resulting in increase of WC, thus increasing the WHtR.

Although this study is confined to a narrow paediatric population, it describes the obesity-related metabolic risk and the inflammatory link between adiposity and adverse metabolic outcomes among the children (8-9 years). The sex-based analysis of data was another strength of this study as sex is an essential factor that could influence adiposity and metabolic and inflammatory risk. Further, this is the first study in Sri Lanka that has reported the cut-offs of WHtR to predict IR and inflammation in this age group of children.

One limitation of the study is that it was not a nationally representative sample, and the findings could not be generalizable for the entire paediatric population of this age group of children in Sri Lanka. However, it could serve as a representative paediatric population of other urban areas of Sri Lanka where a similar socio-economic background is seen. Another limitation is that we did not have data on systolic blood pressure measurements.

Conclusions

The inflammatory and metabolic risks increase with high adiposity. The inflammatory risk increases with adiposity resulting in increase in IR and metabolic abnormalities. The higher IR seen in children with high hs-CRP (>1mg/L) indicates the contributory role of inflammation in the development of IR. Further, the increased metabolic risk in children with high IR indicates the multitude of metabolic derangement arising with IR in this study population. Furthermore, the WHtR cut offs of 0.47 (sensitivity-82.7%) and 0.45 (sensitivity-84.2%) predicting inflammation and 0.48 (sensitivity-90.6%) and 0.46 (sensitivity-90.5%) predicting IR among males and females, respectively, could be used as a cost-effective, simple screening tool to predict IR (>1.7) and inflammation (hs-CRP>1mg/L) for this age group of children.

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