Premature atherosclerosis in Indian children with Juvenile Idiopathic Arthritis: A cross-sectional study

***Suparna Guha[1], Ajanta Haldar[1], Amit Choubey[1], Samar Ranjan Pal[1]**

Sri Lanka Journal of Child Health, 2021; **50**(1): 32-37

Abstract

Background: Commonest cause of chronic arthritis in childhood is Juvenile Idiopathic Arthritis (JIA). Atherosclerosis is a chronic inflammation of the arteries. Both conditions are associated with high inflammatory cytokines. In adults with rheumatoid arthritis, atherosclerosis is a common cause of long term morbidity. Such data in children with JIA, especially from the Indian sub-continent, is lacking.

Objectives*:* To assess the risk of premature atherosclerosis in Indian children with JIA and study associated risk factors.

Method: A cross sectional study was carried out in the department of paediatric rheumatology of Vivekananda Institute of Medical Sciences from 2016 to 2019 involving 42 patients and 21 age and sex matched controls. Premature atherosclerosis was assessed in both cases and controls by measuring bilateral carotid intima thickness (CIMT) using high resolution ultrasonography (USG). For associated risk factors, serum lipid profile and homocysteine levels were measured.

Results: JIA patients had higher CIMT compared to controls (0.5121mm vs 0.429mm, $p<0.001$). Among the subtypes, enthesitis related arthritis showed maximum increase in CIMT (0.5443mm). Considering risk factors, high density lipoprotein (HDL) was lower in cases compared to controls (47.17mg/dl vs 52.20mg/dl, $p<0.02$); homocysteine was more in cases compared to controls (20.76micromol/L vs 13.51micromol/L, $p<0.01$)

Conclusions: Our cohort of Indian children with JIA had a higher CIMT and higher associated risk factors compared to normal healthy children making them more prone to premature atherosclerosis.

DOI: http://dx.doi.org/10.4038/sljch.v50i1.9399

(Key words: Atherosclerosis, CIMT, JIA, homocysteine)

[1]*Vivekananda Institute of Medical Sciences (VIMS), Kolkata India*

**Correspondence:* drs68guha@gmail.com

https://orcid.org/0000-0001-6410-0607

(Received on 12 February 2020: Accepted after revision on 22 March 2020)

The authors declare that there are no conflicts of interest

The study was funded by the VIMS fund.

Introduction

Worldwide prevalence of Juvenile Idiopathic Arthritis (JIA) is 0.07 to 4.1/1000 children and the incidence is 0.008 to 0.226 cases/1000 children[1]. However, exact incidence and prevalence of JIA in India is unknown. Apart from musculoskeletal debility, recently there has been increasing concern regarding the long term outcome of JIA, mainly in the form of cardiovascular disease[2].

In adults with rheumatoid arthritis (RA), cardiovascular disease is about 60% more than the general population[3]. Thus, according to European League against Rheumatism (EULAR) guidelines[4], all adults with long standing RA should have an annual cardiovascular risk assessment. Unlike adults, evidence of long term cardiovascular risk in children with inflammatory arthritis is lacking and there are few guidelines regarding assessment as well as management. Though the risk of premature atherosclerosis in children is already known, in the Indian subcontinent, such data are scarce.

About 40-78% of children with JIA have a chronic course requiring long-term therapy[5]. In fact, more than one third of JIA patients have disease which persists into adulthood[6] and these are the children who are prone to develop atherosclerotic changes. It is very likely that the effects of atherosclerosis in the form of ischaemic heart disease or cerebrovascular events which we see in adults may actually have had its origin in childhood.

Objectives

To assess the risk of premature atherosclerosis in Indian children with JIA and to study associated risk factors.

Method

The study was carried out at the Paediatric Rheumatology Department from February 2016 to June 2019. Of all the 100 JIA children attending the Paediatric Rheumatology Out-Patient Department (OPD) during the study period, 42 cases who

agreed to participate in the study and had a disease duration of more than one year were selected. Twenty one age and sex matched controls were selected from the general paediatric OPD. These were healthy children without any organic complaints who had come for routine check-up. Those with pre-existing cardiovascular risks like obesity, hypertension, diabetes, history of smoking, or a family history of premature stroke or any form of cardiovascular disease or family history of hypertension or diabetes were excluded from the study. All participants had a thorough clinical examination which included measuring body weight, height and blood pressure. Body mass index (BMI) was calculated according to the formula weight in kg/height in m^2.

Laboratory measurements: Fasting (the participants were requested to complete dinner by 8pm, and blood was collected the next morning at 8am) blood was collected by venepuncture for lipid profile, homocysteine (HCYS), serum folate, and vitamin B12 levels. Lipid profile was measured by spectrophotometry and HCYS, folate and B12 assay was done by chemiluminescence immune assay (CLIA). These tests were carried out in the biochemistry department.

Vascular study: Common carotid artery (CCA) intima media thickness (IMT) measurement was done in all participants using B mode ultrasonography (USG; machine specifications being GE Volution S8) with high resolution linear probe (12HZ). Three consecutive longitudinal images of each CCA, 1-2cm proximal to the bifurcation was acquired. The mean value of IMT for right and left CCA was obtained by averaging the 3 measurements of each artery. Finally, the mean carotid IMT of the CCAs was determined and expressed in mm. This was conducted at the radiology department.

Ethical issues: Ethical clearance was obtained from the Ethical Review Committee of Vivekananda Institute of Medical Sciences, Kolkata, India (No: ECR/62/Inst/WB/2013). Written informed consent of the patients and their caregivers was taken.

Statistical analysis: Continuous data are presented as mean ± SD. Continuous variables were checked for normality assumption using Kolmogrov-Smirnov Z test. Coronary intima media thickness (CIMT), homocysteine (HCYS), BMI and cholesterol (CHOL) were not normally distributed. Independent samples t-test was used to compare the normal continuous variables between patients with JIA and healthy controls, while Mann Whitney U test was carried out for comparing non normal continuous variables. Chi-square test was used to compare categorical variables. Kruskal Wallis test was conducted for comparison of factors among different JIA types for non-normal data and post hoc analysis was done using Dunn Kruskal Wallis Multiple Comparison. One way ANOVA was carried out in case the data could be assumed to be from the normal population. P-value (one tailed and two tailed) less than 0.05 was considered to be significant. The analysis was done using R programming language.

Results

The 42 JIA patients were categorised in accordance with ILAR criteria. Of them, 15 were systemic onset (SOJIA), 14 were enthesitis related (ERA), 9 were polyarticular onset (POLYJIA), one each were psoriatic and undifferentiated and 2 were oligo articular (OJIA). All patients with ERA had HLA-B27 done.

From Table 1, it can be seen that the maximum number of cases were in the adolescent age group of 12-16 years. Among the 42 cases, 19 were female and 23 were male. The control group had 10 females and 11 males. All our subjects had a disease duration of more than 1 year when we first encountered them in our OPD.

Table 1: ***Distribution of Juvenile idiopathic arthritis (JIA) according to age***

Age group (years)	ERA	POLYJIA	SOJIA	Total
2-6	-	-	06	06
7-11	03	03	03	09
12-16	11	06	06	23
Total	14	09	15	38

ERA: enthesitis related arthritis, POLYJIA: polyarticular onset JIA, SOJIA: systemic onset JIA

Disease activity was evaluated by active tender swollen joint count, presence of systemic features, ESR and general health. Of these 42 patients, 22 had active disease at the time of recruitment. All these cases were receiving treatment at the time of recruitment; 12 cases were on methotrexate, 5 were on leflunamide, 7 were on steroids and 3 were on biologic agents.

Comparison of cases and controls

The average age of JIA patients was 11.14±3.66 years and the average age for the control group was

11.65±3.01 years. The mean CIMT of JIA subjects was 0.512±0.08mm while that of controls was 0.4295±0.05mm. This was statistically significant (p<0.001).

The mean HCYS in JIA subjects was 20.76±10.66 micromol/L while that of controls was 13.51±2.18 micromol/L. The Wilcoxon rank sum test gave a p-value of <0.01 which is statistically significant. Regarding vitamin B12 and folic acid both groups had comparable levels.

In lipid profile, fasting total cholesterol and HDL levels were performed. Regarding total cholesterol, the p-value was 0.2084 and did not show any statistical significance. Mean HDL levels in JIA patients was 47.17±7.87 mg/dl while controls showed a mean value of 52.20±5.98 mg/dl which was statistically significant (p<0.01)

Table 2: ***Comparison of Juvenile idiopathic arthritis (JIA) cases and controls***

Parameters	JIA *(n=42)* (Mean ± SD)	Control *(n=21)* (Mean ± SD)	p-value
Age (years)	11.14 ± 3.36	11.65 ± 3.01	0.947
Female	19 ± 45.24	10 ± 47.62	0.8581
Body mass index (kg/m^2)	17.93 ± 4.67	17.15±3.89	0.6992
Coronary intima media thickness (mm)	0.5121 ± 0.08	0.4295 ± 0.05	*<0.001*
Homocysteine (micromol/L)	20.76 ± 10.66	13.51 ± 2.18	*<0.01*
Cholesterol (mg/dl)	147.33 ± 24.81	139.65 ± 19.83	0.2084
High density lipoproteins (mg/dl)	47.17 ± 7.87	52.20 ± 5.08	*<0.02*

Comparison of the JIA sub types

For analysis among different subtypes of JIA, only 3 groups, SOJIA, ERA and PJIA types were chosen as other types had very small numbers (1 each of psoriatic and undifferentiated and 2 OJIA). Among the various JIA subtypes, CIMT was mostly increased in ERA. The post hoc testing using Dunn Kruskal Wallis multiple comparison showed that among the chosen variables, CIMT differs significantly among the various subtypes of JIA. For all other factors there were no significant differences among the various subtypes.

Table 3: ***Comparison of various Juvenile idiopathic arthritis (JIA) subtypes***

Parameter	ERA (Mean ± SD)	POLYJIA (Mean ± SD)	SOJIA (Mean ± SD)	p-value
Body mass index (kg/m^2)	18.99 ± 5.85	17.53 ± 2.90	16.58 ± 3.89	0.509
Coronary intima media thickness (mm)	0.5443 ± 0.07	0.5233 ± 0.09	0.4640 ± 0.08	*0.027*
Homocysteine (micromol/L)	22.95 ± 13.98	21.17 ± 6.19	20.59 ± 9.50	0.9144
Total cholesterol (mg/dl)	145.64 ± 14.49	137.67 ± 27.46	160.00 ± 28.06	0.07
High density lipoproteins (mg/dl)	45.48 ± 7.86	45.78 ± 5.54	49.47 ± 9.67	0.382

ERA: enthesitis related arthritis, POLYJIA: polyarticular onset JIA, SOJIA: systemic onset JIA

Table 4: ***Comparison of the present study with other studies***

Parameter	Urban *et al*	Vlahos *et al* (Mean ± SD)	Present study (Mean ± SD)
Total cholesterol (mg/dl)	-	164 ± 29	147.33 ± 24.81
High density lipoproteins (mg/dl)	-	51 ± 12	47.17 ±7.87
Homocysteine (micromol/L)	8.2	-	20.76 ±10.66
Coronary intima media thickness (mm)	0.43	0.46 ± 0.03	0.5121 ± 0.08

Discussion

The traditional risk factors for atherosclerosis include diabetes, dyslipidaemia, obesity, hypertension and tobacco smoking. It was in 1999 that Russel Ross first pointed out his theory of atherosclerosis being an inflammatory process[7]. The earliest changes in atherosclerosis is a functional and structural abnormality of the endothelial vessel wall[12]. This process is initiated by an inflammatory reaction involving lymphocytes, macrophages and a host of cytokines like IL-6, IL-16, IL-8 and tumour necrosis factor (TNF). The pathology is somewhat similar to that seen in other chronic inflammatory diseases like connective tissue disorders, RA and JIA where an interplay of pro-inflammatory cytokines and inflammatory cells result in expression of various clinical features. Cardiovascular disease is one of the main causes of deaths in adults with RA[8,9]. In adults, CIMT in patients with RA is significantly increased in comparison to healthy controls[13].

In children with JIA, though there are reports of cardiovascular involvement in the form of

premature atherosclerosis[14,15,16], studies from the India are lacking. Early atherosclerotic changes in JIA subjects were demonstrated in a Polish study by Pietrewicz *et al*[14] and in a Greek study by Vlahos *et al*[15]. While in the Polish study the authors observed that CIMT was significantly increased in the PJIA group (p=0.003), the Greek study demonstrated that the SOJIA patients had a significant increase in the CIMT (p<0.001) compared to the other groups. The findings in the above studies are in contrast to our observation of a significant increase of CIMT in the ERA group. This relatively greater increase in CIMT in our ERA cohort can perhaps be attributed to the higher age group of this cohort, the greater disease duration and the longer period of active disease
.

Apart from the CIMT, which is a non-invasive surrogate marker of premature atherosclerosis, this study also included assessment of some risk factors. Among the newer risk factors for atherosclerosis, homocysteine (HCYS) is an emerging offender. It is a sulphur containing amino acid, increased concentration of which causes increased oxidation of LDL side chains which leads to increased free radical formation and causing endothelial damage subsequently increasing the risk of atherosclerosis[10,11,17]. The risk of homocysteine and vascular disease has been well cited[18-21]. Vitamin B12 and folic acid are essential for the metabolism of HCYS and a deficiency of these 2 elements may also cause a rise in HCYS levels. In our current study, HCYS levels were significantly increased (p<0.01) in JIA patients in spite of normal B12 and folate levels. From this study it can be seen that Indian JIA children have increased HCYS levels which may be a potential risk factor for premature atherosclerosis.

In adults with RA, evidence for dyslipidaemia as a risk factor for atherosclerosis has also been shown. However, in studies conducted on JIA patients, there have been conflicting reports regarding lipid levels. While our study and a study by Gonclaves *et al*[20] have shown a significant derangement of lipid function, studies by Vlahos *et al*[16] did not demonstrate such association. In our study, though levels of both total cholesterol and HDL were deranged, significant differences between cases and controls were seen only with HDL levels.

Cardiovascular risk in children with JIA is a lesser known entity. This is perhaps the first study from India which could demonstrate atherosclerosis as a long term complication of JIA. Further studies may be needed in the future to study the progress of atherosclerosis in these subjects and corroborate our findings. Since JIA children possess risk factors, which make them prone to premature atherosclerosis, they may also need cardiovascular monitoring and CIMT measurement may help to predict premature atherosclerosis in such patients.

Conclusions

Our cohort of Indian children with JIA had a higher CIMT and higher associated risk factors compared to normal healthy children making them more prone to premature atherosclerosis.

Acknowledgements

We acknowledge the help received from Ms Snigdha Pain, who helped in our statistical analysis, Ms Kakoli Ghosh, our research scholar and Dr. Mitali Das, OPD medical officer in rheumatology.

References

1. Goldmuntz EA, White PH. Juvenile idiopathic arthritis: a review for the paediatrician. *Pediatrics in Review* 2006; **27**(4):e24-32. https://doi.org/10.1542/pir.27-4-e24 PMid: 16581950

2. Avina–Zubieta JA, Choi HK, Sadatsafavi M, Etminan M, Esdaile JM, Lacalle D. Risk of cardiovascular mortality in patients with rheumatoid arthritis: a meta-analysis of observational studies. *Arthritis and Rheumatism* 2008; **59**(12):1690-7. https://doi.org/10.1002/art.24092 PMid: 19035419

3. Turesson A, Jaccobson L. Increased incidence of cardiovascular disease with rheumatoid arthritis: results from a community based study. *Annals of Rheumatic Diseases* 2004; **63**(8):952-5. https://doi.org/10.1136/ard.2003.018101 PMid: 15051620 PMCid: PMC1755101

4. Peters MJ, Symmons DP, McCarey D, Dijkmans BA, Nicola P, Kvien TK, et al. EULAR evidence based recommendations for cardiovascular risk management in patients with rheumatoid arthritis and other forms of inflammatory arthritis. *Annals of Rheumatic Diseases* 2010; **69**(2):325-31. https://doi.org/10.1136/ard.2009.113696 PMid: 19773290

5. Nordal E, Zak M, Alto K, Bernston L, Fasth A, Herlin T *et al.* Ongoing disease activity and changing categories in a long term Nordic cohort study of Juvenile

Idiopathic Arthritis. *Arthritis and Rheumatism* 2011; **63**(9):2809-18. https://doi.org/10.1002/art.30426 PMid: 21560116

6. Packham JC, Hall MA. Long-term follow-up of 246 adults with juvenile idiopathic arthritis: functional outcome. *Rheumatology* 2002; **41**(12):1428-35. https://doi.org/10.1093/rheumatology/41.12.1428 PMid: 12468825

7. Ross R. Mechanisms of disease: atherosclerosis –an inflammatory disease. *New England Journal of Medicine* 1999; **340**:115-26. https://doi.org/10.1056/NEJM199901143400207 PMid: 9887164

8. Mahmood SS, Levy D, Vasan RS, Wang TJ. The Framingham Heart Study and the epidemiology of cardiovascular disease: a historical perspective. *Lancet* 2014; **383**(9921): 999-1008. https://doi.org/10.1016/S01406736(13)61752-3

9. Solomon DH, Karlson EW, Rimm EB, Cannuscio CC, Mandl LA, Manson JE, *et al.* Cardiovascular morbidity and mortality in women diagnosed with rheumatoid arthritis. *Circulation* 2003; **107**(9):1303-7. https://doi.org/10.1161/01.CIR.0000054612.26458.B2 PMid: 12628952

10. Urban M, Pietrewicz E, Gorska A, Glowinska B. Lipid and homocysteine levels in Juvenile Idiopathic Arthritis. *Pol Merkur Lekarski* 2004; **17**: 235-8.

11. Hankey GJ, Eikeboom JW. Homocysteine and vascular disease. *Lancet* 1999; **354**(9176): 407-13. https://doi.org/10.1016/S01406736(98)11058-9

12. Davignon J, Ganz P. Role of endothelial dysfunction in atherosclerosis. *Circulation* 2014; **109**(Suppl 1): 27-32. https://doi.org/10.1161/01.CIR.0000131515.03336.f8 PMid: 15198963

13. AM.Sijl Van, MJ.Peters et al.Carotid-intima Media Thickness in rheumatoid arthritis compared to control subjects: a ameta-analysis. *Seminars in Arthritis and Rheumatism* 2011; **40**(5): 389-97 https://doi.org/10.1016/j.semarthrit.2010.06.006 PMid: 20889191

14. Pietrewicz E, Urban M. Early atherosclerosis changes in children with juvenile idiopathic arthritis. *Pol Merkur Lekarski* 2007; **22**(129):211-4.

15. Vlahos AP, Theocharis P, Bechlioulis A, Vakalis K, Alfantaki S, et al. Changes in vascular function and structure in Juvenile Idiopathic Arthritis. *Arthritis Care and Research* 2011; **63**(12):1736-44. https://doi.org/10.1002/acr.20613 PMid: 21905249

16. Amar A. Hanam, Elhewala A.I. Subclinical atherosclerosis in Egyptian children with JIA: assessment of the risk factors. *International Journal of Advanced Research* 2015; **3**(11): 1138-50

17. Thmbyarajah J, Towned JN. Homocysteine and atherothrombosis: Mechanisms for injury. *European Heart Journal* 2000; **21**(12): 967-74. https://doi.org/10.1053/euhj.1999.1914 PMid: 10901508

18. Del Rincon I, Williams K, Stern MP, Freeman GL, Escalante A. High incidence of cardiovascular events in rheumatoid arthritis cohort not explained by traditional cardiac risk factors. *Arthritis and Rheumatism* 2001; **44**:2737-45. https://doi.org/10.1002/15290131(200112)44:12<2737::AID-ART460>3.0.CO;2-#

19. Huemer M, Fodinger M, Huemer C, Sailer-Hock M, Falger J, Rettenbacher A, *et al*. Hyper-homocysteinemia in children with juvenile idiopathic arthritis is not influenced by methotrexate treatment and folic acid supplementation: a pilot study. *Clinical and Experimental Rheumatology* 2003; **21**(2):249-55.

20. Gonclaves M, Almeida VD, Guerra-Shinohara EM, Galdieri LC, Len CA, Hilario MOE. Homocysteine and lipid profile in children with Juvenile Idiopathic Arthritis. *Pediatric Rheumatology* 2007; **5**: Article No. 2.

https://doi.org/10.1186/1546-0096-5-2
PMid: 17550630 PMCid: PMC1869026

21. Selhub J, Jacques PF, Bostom AG, D'Agostino RB, Wilson PW, Belanger AJ, *et al*. Association between plasma homocysteine concentration and extra-cranial carotid artery stenosis. *New England Journal of Medicine* 1995; **332**(5):286-91. https://doi.org/10.1056/NEJM199502023320502
PMid: 7816063