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IP Indian Journal of Clinical and Experimental Dermatology

Journal homepage: www.ijced.org/

Original Research Article

Clinicoepidemiological study of acanthosis nigricans in pre and primary school children and its association with body mass index, waist circumference and insulin resistance

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ARTICLE INFO

Article history:

Received 04-08-2022

Accepted 09-08-2022

Available online 29-09-2022

Keywords:

Acanthosis nigricans (AN)

BMI

WC

HOMA2IR

insulin resistance (IR)

ABSTRACT

Introduction: Acanthosis nigricans is characterised by hyperpigmentation and velvet like thickening of skin. It is seen symmetrically involving the neck, axilla, groins, antecubital and popliteal fossae, umbilical, perianal areas and in advanced conditions, even dorsum of hands and fingers.

Objectives: To evaluate the clinical features and epidemiological factors of acanthosis nigricans in pre and primary school children and to evaluate the association of acanthosis nigricans with body mass index, waist circumference and insulin resistance.

Materials and Methods: A total of 100 children with acanthosis nigricans meeting defined inclusion and exclusion criteria were enrolled in this cross-sectional study after taking an informed consent and approval of institutional ethical committee. All parents of children with acanthosis nigricans were subjected to a detailed history based on a questionnaire. A thorough clinical examination was done to study acanthosis nigricans. Blood samples were collected and fasting serum insulin and fasting blood sugar were determined.

Results: Most of the children were boys (M:F=1.3:1) and mean age was 6.66±1.99 years. Twenty four percent were normal weight, 28% were overweight and 48% were obese children. On examination, neck was the commonest site of acanthosis nigricans. Grade 4 neck severity (p=0.0014) and grade 3 neck texture (P=0.0198) of acanthosis nigricans were significantly associated with Insulin resistance (HOMA2-IR). Twenty eight percent of normal weight, 42.8% of overweight and 75% of obese children had insulin resistance.

Conclusion: Acanthosis nigricans was associated with insulin resistance in normal, overweight and obese children. Hence, we would like to propose that all the children with acanthosis nigricans must be screened for Insulin resistance irrespective of body mass index. With this evidence of Insulin resistance, guidelines can be given to the parents about life style modification which will help in preventing the onset of diabetes mellitus later and thereby, improving the quality of life of the child.

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1. Introduction

Acanthosis nigricans is characterized by dark brown and velvety thickening of the skin. It is symmetrically distributed on the neck, axillae, antecubital fossae, popliteal

fossae, groin folds and face.¹ In children, acanthosis nigricans mainly affects the neck (93-99%) followed by axillary area (73%).² It is a common condition associated with obesity, which in turn is accompanied by hyperinsulinemia and diabetes mellitus.³ Hence, acanthosis nigricans is aptly a marker of insulin resistance and hyperinsulinemia, with or without diabetes mellitus.

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In a study done by Mrinal gupta, among 100 obese children the most common cutaneous manifestation was found to be acanthosis nigricans (42%).⁴ In another study done by Shalitin S et al in Israel, among 234 obese children and adolescents 56.6% manifested with acanthosis nigricans and 81% were diagnosed with insulin resistance.⁵ In recent years, there is alarming increase in prevalence of obesity in children due to the adoption of western dietary habits, lack of physical activity and other health habits.⁶ Hence, more number of cases of childhood obesity and acanthosis nigricans can be seen nowadays. Studies indicate that children with acanthosis nigricans have higher levels of basal and glucose-stimulated insulin compared with obese children without acanthosis nigricans, suggesting an association of acanthosis nigricans with hyperinsulinemia independent of body mass index.⁷

Obesity is a proven risk factor for diabetes mellitus, a multisystem chronic disorder.⁸ Early detection of acanthosis nigricans aids in recognition of insulin resistance and thereof diabetes mellitus. Institution of life style intervention can prevent or delay the onset of type 2 diabetes by as much as 58 %.⁹

Since there is paucity of studies on acanthosis nigricans in Indian children, we mainly aim to assess the pattern of acanthosis nigricans in preschool and primary school children and its association with various parameters like body mass index, waist circumference and insulin resistance. With this evidence, guidelines can be given to the parents of children with acanthosis nigricans about life style modification, reducing the risk factors for obesity and thereby helping in reducing major complications of insulin resistance in future such as diabetes mellitus, cardiovascular diseases, dyslipidemia and thereby improving the quality of life of the child.

2. Materials and Methods

This study was conducted in anganwadis, preschools and primary schools which come under the field practice area of Adichunchanagiri Institute of Medical Sciences, B.G. Nagara, Mandya District.

2.1. Sample size

100 Cases.

2.2. Sample procedure

Cross-sectional study.

2.3. Study duration

December 2019 to June 2021.

2.4. Inclusion criteria

1. Parents/guardians giving an informed consent for enrolling their child in the study
2. Preschool and primary school children between the age group of 3-10 years with acanthosis nigricans, diagnosed clinically.

2.5. Exclusion criteria

1. Parents/guardians not willing to enroll their child in the study.
2. Children diagnosed with primary metabolic diseases like primary dyslipidemia, type I diabetes mellitus and thyroid disorders.
3. Children on medication that causes drug induced acanthosis nigricans like insulin, nicotinic acid and systemic steroids.

2.6. Procedure of the study

2.6.1. Method of collection of data

All preschool and primary school children in field practice area of B.G. Nagara and satisfying aforementioned inclusion and exclusion criteria were recruited in the study. Approval was obtained from institutional ethical committee and written informed consent was taken from the parent or guardian of child with acanthosis nigricans. Patient with acanthosis nigricans was considered in the study if they had dermatosis characterized by velvety, papillomatosis, brownish to black discoloration, thickened, hyperpigmented plaques, typically of neck and intertriginous area.

2.6.2. Clinical history

A detailed history was obtained from parent/guardian of child with acanthosis nigricans based on standard prepared questionnaire and emphasis was laid on history of darkening of skin, duration, onset, initial site and distribution. Personal history suggestive of juvenile diabetes and overweight was considered. Family history of acanthosis nigricans, obesity, cardiovascular disease and diabetes mellitus was documented.

2.6.3. Clinical examination

A thorough clinical examination was done. Presence of pallor, icterus, cyanosis, clubbing, edema and lymphadenopathy was noted. Anthropometric measurement was done based on standard protocol recommended by WHO.^{10,11}

2.6.4. Measurement protocols

2.6.4.1. Height. Measured against vertical board with an attached metric rule and bringing horizontal head board in contact with the uppermost point on the head. Recorded in bare foot, full erect position and deep inspiration.

2.6.4.2. Weight. Recorded without foot wear and light clothes on ISI (Indian standards institute) certified weighing machine to the nearest of 100gm.

2.6.4.3. Body mass index. Calculated as weight in kg / height in m² (kilogram / meters²). The scoring was done according to WHO growth charts.

In children overweight is defined as BMI between the 85th and 94th percentile for age and gender, while obese is defined as BMI more than or equal to 95th percentile for age and gender.

2.6.4.4. Waist circumference. Will be measured at the midpoint between the lower margin of the last palpable rib and the top of the iliac crest, using stretch-resistant tape that provides a constant 100g tension. It was measured twice, with a maximum variation of 1cm and the average was taken.

2.6.5. Cutaneous examination

Diagnosis of acanthosis nigricans was based on clinical morphology. Its presence in different sites such as axillae, posterior neck fold, flexor surface of upper and lower extremities, umbilicus, groin, inframammary folds, face, and perioral, perianal surface, periorbital, maxillary, periocular and mucosal surface of conjunctivae, lips, oral cavity and vulva was noted. Acanthosis nigricans over neck, axilla and other sites was scored quantitatively according to Bruke's scale.¹² Systemic examination was also done.

2.6.6. Investigations

Blood samples were collected from all enrolled patients and the following investigations was performed.

1. Fasting serum insulin levels.
2. Fasting blood sugar levels (FBS).

2.7. Diagnosis of insulin resistance

HOMA2-IR (Homeostasis model assessment of insulin resistance) is used to assess the association of acanthosis nigricans and insulin resistance. HOMA-IR calculator V.2.2.3 was used to assess the insulin resistance. A value above 1.8 was considered as insulin resistance.¹³

2.8. Method followed in the investigation

Venous plasma glucose will be measured by glucose oxidase method. Fasting blood glucose more than 110mg% will be considered raised.

Serum insulin levels will be measured by Microplate Chemiluminescence Assay using the instrument Alpha prime light. The normal reference range for serum insulin was 5–25 μ IU/ml.

The Statistical software namely SAS 9.2, SPSS 15.0, Stata 10.1, MedCalc 9.0.1, Systat 12.0 and R environment

ver.2.11.1 were used for the analysis of the means, medians and standard deviations. Student t test (two tailed, independent) has been used to calculate the statistical significance. Chi-square/ Fisher Exact test has been used to find the significance of study parameters on categorical scale between two or more groups.

3. Results

3.1. Age distribution of children with AN

Out of 100 children with acanthosis nigricans, 29% of children were in the age group 3-5 years and 71% were in the age group 6-10 years. Mean age duration \pm standard deviation was found to be 6.66 \pm 1.99.

3.2. Gender distribution of children with AN

Boys (58%) outnumbered girls (42%). Male: Female ratio is 1.3:1.

Above table suggests that majority of children (50%) had duration of darkening of skin of less than 1 year. Majority of them (88%) had an insidious onset. Neck was the commonest initial site of darkening (87%), followed by axilla (4%), periorbital region (3%), knuckles (2%), and inframammary regions (2%) and groin (2%).

3.3. Family history of acanthosis nigricans and other medical illness

22% of children had a positive family history of acanthosis nigricans, 25% had a family history of diabetes mellitus, 16% had a family history of hypertension and 3% had a family history of cardiovascular disease.

3.4. BMI of the children with AN

Out of 100 children, 48% were obese, 28% were overweight and 24% had normal weight.

3.5. Distribution of waist circumference of children with AN

According to the waist circumference, 51% of the children had central obesity.

3.6. Distribution of children according to central obesity

32.1% of overweight children and 87.5% of obese children had central obesity

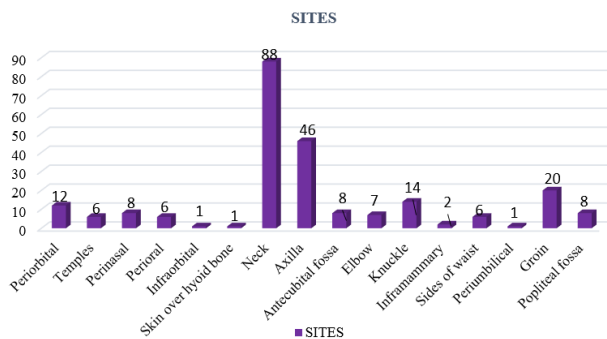
Neck (88%) was the most commonest site of acanthosis nigricans followed by axilla (46%); groin (20%); knuckles (14%); periorbital region (12%); perinasal region, popliteal fossa and antecubital fossa (8%) each; elbow (7%); temples, sides of waist and perioral (6%) each; inframammary (2%); skin over hyoid bone, periumbilical and infraorbital region

Table 1: History of darkening of skin with duration, onset of darkening of skin and initial site of darkening

Parameters		Number of children	Percentage (%)
History of darkening of skin with duration	Not noticed	24	24.0
	<1year	50	50.0
	>1year	26	26.0
	Total	100	100.0
Onset	Insidious	88	88.0
	Acute	12	12.0
	Total	100	100.0
Initial site of darkening	Neck	87	87.0
	Periorbital region	3	3.0
	Knuckle	2	2.0
	Infra-mammary Region	2	2.0
	Axilla	4	4.0
	Groin	2	2.0
	Total	100	100.0

Table 2: Grading of neck severity, neck texture and axillary severity and their association with insulin resistance

G grade	No. of children	Insulin resistance (HOMA2-IR>1.8)	Non-insulin resistance (HOMA2-IR<1.8)	P value
Neck severity				
Grade 0	9(9%)	2(3.8%)	7(14.5%)	0.65
Grade 1	10(10%)	2(3.8%)	8(16.6%)	0.07
Grade 2	20(20%)	7(13.4%)	13(27.08%)	0.14
Grade 3	35(35%)	20(38.4%)	15(31.2%)	0.58
Grade 4	26(26%)	21(40.3)	5(10.4%)	0.0014
Neck texture				
Grade 0	12(12%)	5(9.6%)	7(14.5%)	0.648
Grade 1	13(13%)	4(7.6%)	9(18.7%)	0.178
Grade 2	44(44%)	21(40.3%)	23(47.9%)	0.5
Grade 3	31(31%)	22(42.3%)	9(18.7%)	0.0198
Axillary severity				
Grade 0	40(40%)	18(34.6%)	22(45.8%)	0.3476
Grade 1	12(12%)	8(15.3%)	4(8.3%)	0.4371
Grade 2	20(20%)	11(21.1%)	9(18.7%)	0.9600
Grade 3	14(14%)	7(13.4%)	7(14.5%)	0.8990
Grade 4	14(14%)	8(15.3%)	6(12.5%)	0.8990



Graph 1: Sites having acanthosis nigricans

(1%) each.

3.7. Neck severity

Grade 4 neck severity was more positively associated with Insulin resistance (HOMA2-IR) and was found to be statistically significant (p=0.0014).

3.8. Neck texture

Grade 3 neck texture was more positively associated with Insulin resistance (HOMA2-IR) and was found to be statistically significant p=0.0198.

3.9. Axillary severity

Axillary severity was statistically not associated with Insulin resistance (HOMA-2IR).



Fig. 1: Neck severity 2 [acanthosis nigricans limited to the base of the skull, does not extend to the lateral margins of the neck (usually <3 inches in breadth)]



Fig. 2: Neck severity 3 [acanthosis nigricans extending to the lateral margins of the neck (posterior border of the sternocleidomastoid) (usually 3-6 inches), should not be visible when the participant is viewed from the front]



Fig. 3: Neck severity 4 [acanthosis nigricans extending anteriorly (>6 inches), visible when the participant is viewed from the front]

3.10. Neck severity(Figures 1, 2 and 3)

3.11. Neck texture(Figures 4, 5 and 6)



Fig. 4: Neck texture 1 [rough to touch: clearly differentiated from normal skin]

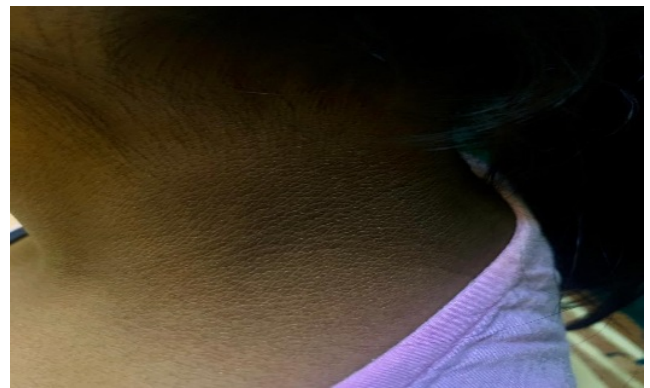


Fig. 5: Neck texture 2 [coarseness can be observed visually, portions of the skin clearly raised above other areas]

3.12. Axilla severity(Figures 7, 8, 9 and 10)

The FSI ($p=0.0025$) and HOMA2-IR ($p=0.0028$) values were significantly higher in overweight compared to normal weight AN children.

The FBS ($p=0.0095$), FSI ($p=0.0001$) and HOMA2-IR ($p=0.0001$) values were significantly higher in obese compared to normal weight AN children

The FBS ($p=0.0218$), FSI ($p=0.0011$) and HOMA2-IR ($p=0.01059$) values were significantly higher in obese compared to overweight AN children.

The study showed significant difference between IR positive and IR negative children in terms of BMI ($p=0.000192$), fasting blood glucose ($p=<0.00001$),

Table 3: Mean levels of fasting blood sugar, fasting serum insulin and HOMA IR levels in normal, overweight and obese children.

Parameters	Normal (n=24)	Overweight (n=28)	Obese (n=48)	Comparison between normal and overweight	Comparison between normal and obese	Comparison between obese and overweight
Fasting blood sugar (mg/dL), mean±SD	84.4583±4.9165	84.9643±8.7156	90.4167±10.353	0.8021	0.0095	0.0218
Fasting Serum insulin level (μIU/mL), mean±SD	10.9779±2.57	13.7136±3.4739	17.0835±4.5441	0.0025	0.0001	0.0011
HOMA IR mean±SD	1.3867±0.3288	1.7289±0.4367	2.1865±0.6084	0.0028	0.0001	0.0008
Insulin resistance (HOMA2-IR>1.8), n(%)	4 (28.5%)	12(42.8%)	36 (75%)	0.0821	<0.00001	0.01059

Table 4: Demographic and biochemical features of IR positive & IR negative children.

Parameters	IR negative	IR positive	p value
Age	6.6±2.2	6.71±1.80	.394569
Gender	27:21	21:31	.112617
BMI	18.1±1.95	19.77±2.05	.000192
Fasting glucose	83.35±6.95	91.25±9.52	<.00001
Fasting insulin	11±2.05189	18.06 ±3.54	<.00001
Homa IR	1.4 ± 0.27	2.32 ± 0.48	<.00001
Waist circumference	65.27 ± 9.61	67.65 ± 6.90	.10063

**Fig. 6:** Neck texture 3 [extremely coarse: “hills and valleys” observable on visual examination]

fasting serum insulin levels ($p < 0.00001$) and HOMA-IR ($p = 0.00001$), which was statistically significant.

4. Discussion

Acanthosis nigricans is characterized by hyperpigmentation and velvet like thickening of skin. It is seen symmetrically involving the neck, axilla, groins, antecubital and popliteal fossae, umbilical, perianal areas and in advanced conditions, even dorsum of hands and fingers. It is strongly associated with obesity, which in turn is accompanied by

**Fig. 7:** Axilla severity 2 [acanthosis nigricans localized to the central portion of the axilla, may have gone unnoticed by the participant]



Fig. 8: Axilla severity 3 [acanthosis nigricans involving the entire axillary fossa, but not visible when the arm is against the participant's side]



Fig. 9: Axilla severity 4 [acanthosis nigricans involving entire axilla and extending to front and back]



Fig. 10: Axilla severity 4 [acanthosis nigricans visible from front in the unclothed participant when the arm is adducted].

hyperinsulinemia and development of diabetes mellitus. Hence, acanthosis nigricans is aptly a marker of insulin resistance and hyperinsulinemia, with or without diabetes mellitus.

Since there is paucity of studies on acanthosis nigricans in Indian children, we mainly aim to assess the pattern of acanthosis nigricans in preschool and primary school children and its association with various parameters like body mass index, waist circumference and insulin resistance.

4.1. Age distribution

In the present study out of 100 children with acanthosis nigricans, majority of children (71) were in age group 6-10years and 29 children were in age group 3-5 years. There were no similar studies to compare our observation.

4.2. Gender distribution

In the present study, boys outnumbered girls. (Male: female=1.3:1)

In contrary to our observation, in a study done kluczynik CEN et al² (M: F=1:1.9) and Nithun TM et al¹⁴ girls outnumbered boys

4.3. Duration of darkening of skin

Of the 100 children studied, 24 had not noticed darkening of skin. 50 children had noticed darkening within < 1year and 26 had noticed it for more than 1 year.

4.4. Onset

Of the 100 children studied, 88 had an insidious onset and 12 had an acute onset (<2months).

4.5. Initial site of darkening

Most of the children noticed darkening of skin first over the neck (87). Rest of them had noticed over axillary region (4), periorbital (3), inframammary region (2), knuckle (2) and groin (2).

4.6. Family history

In the present study, there was a positive family history of acanthosis nigricans in 22, positive family history diabetes mellitus in 25, positive family history hypertension in 16 and positive family history cardiovascular disease in 3 children. Whereas in a study done by Nithun TM et al fifteen patients had positive family history of acanthosis nigricans and 13 had positive family history diabetes mellitus.¹⁴

4.7. BMI assessment of children with acanthosis nigricans

In the present study out of 100 children with acanthosis nigricans, 48% were obese, 28% were overweight and 24% were normal. Whereas in a study done by Lopez- Alvarenga JC et al in 670 children 65.3% were obese, 53.1% were overweight and 10.6% were normal.¹⁵

4.8. Distribution of children according to central obesity

In the present study, 9(32.1%) overweight children and 42(87.5%) obese children had central obesity which was comparable to Hirschler et al¹⁶ study.

4.9. Sites having acanthosis nigricans on examination

Percentage of children with acanthosis nigricans over neck in the present study (88%) is comparable with Nithun TM et al (90%)¹⁴ study.

Percentage of children with acanthosis nigricans in axilla in the present study (46%) is less than that observed by Nithun TM et al(80%). Percentage of children with acanthosis nigricans in groin in the present study (20%) is less than that observed by Nithun TM et al (58.3%). Percentage of children with acanthosis nigricans in inframammary region in the present study (2%) is less than that observed by Nithun TM et al(5%).¹⁴

Percentage of children with acanthosis nigricans in knuckle in the present study (14%) is more than that observed by Nithun TM et al(3.3%).¹⁴

The additional sites involved in the present study include periorbital region (12%); perinasal region, popliteal fossa and antecubital fossa (8%) each; elbow (7%); temples, sides of waist and perioral (6%) each; skin over hyoid bone, periumbilical and infraorbital region (1%) each.

4.10. Distribution of Mean levels of fasting blood sugar, fasting serum insulin and HOMA IR levels in normal, obese and overweight children

Mean FBS, FSI and HOMA-IR in the study done by Nithun TM et al in children was higher compared to the present study.[21] In the present study 28.6% of normal weight, 50% of overweight and 81.6% of obese had insulin resistance which was comparable with Nithun TM et al study.¹⁴

In a study done by Nithun TM et al in children and youth, FSI and HOMA2-IR values were significantly higher in obese compared to normal weight AN individuals ($p=0.0001$). Whereas in the present study, FBS, FSI AND HOMA2-IR values were significantly higher in obese compared to normal weight AN individuals ($p=0.0095$ for FBS; $p=0.0001$ for FSI and HOMA2-IR).¹⁴

In a study done by Nithun TM et al, FSI and HOMA2-IR values were significantly higher in obese compared to overweight AN individuals ($p=0.08$ for FSI; $p=0.014$ for HOMA2-IR). Whereas in the present study, FBS, FSI AND HOMA2-IR values were significantly higher in obese compared to overweight AN individuals (FBS, $p=0.0095$), (FSI and HOMA2-IR, $p=0.0001$).¹⁴

However, in the present study FSI and HOMA2-IR values were significantly higher in overweight compared to normal AN individuals ($p=0.0025$ for FSI; $p=0.0028$ for HOMA2-IR). Whereas in a study done by Nithun TM et al, there was no significant difference was observed between normal and overweight.¹⁴

4.11. Grading of Acanthosis nigricans and its association with insulin resistance

4.11.1. Neck severity

In the present study, grade 3 and grade 4 neck severity was seen in 35(35%) and 26(26%) patients, of which 38.4% and 40.3% patients had IR, respectively. Grade 4 neck severity was statistically associated with HOMA-IR, $p=0.0014$. Whereas in a study done by Nithun TM et al, grade 3 and grade 4 neck severity was seen in 18 (30%) and 12 (20%) cases, of which 61.1% and 83.3% cases had IR, respectively. Grade 4 neck severity was statistically associated with HOMA-IR, $p=0.007$.¹⁴

4.11.2. Neck texture

In the present study, grade 3 was seen in 31(31%) patients, of which 42.3% children had IR and grade 3 neck texture was statistically associated with HOMA-IR, $P=0.0198$. Whereas in a study done by Nithun TM et al, grade 3 was seen in 14(23%), and 12(85.7%) of them showed IR. Grade 3 neck texture was statistically associated with HOMA-IR, $p=0.04$.¹⁴

4.11.3. Axillary severity

In the present study, grade 3 and grade 4 neck severity was seen in 14(14%) patients each, of which 13.4% and 15.3% patients had IR, respectively. Axilla texture is statistically not associated with HOMA-IR.

In a study done by Nithun TM et al, grade 3 and grade 4 neck severity was seen in 12(20%) patients each, of which 24.11% and 37.9% cases had IR, respectively. Grade 4 axillary severity was statistically associated with HOMA-IR, $p=0.002$.¹⁴

4.11.4. Demographic and biochemical features in IR negative and IR positive children

In the present study fasting glucose, fasting insulin and HOMA-IR were higher in IR positive children than IR negative and were found to be statistically significant ($p<0.0001$). Whereas in a study done by Aswani R et al, fasting glucose, fasting serum insulin and HOMA-IR were higher in IR positive children than IR negative but only fasting insulin and HOMA-IR were found to be statistically significant ($p<0.0001$).¹⁷

5. Conclusion

1. Acanthosis nigricans predominantly involved boys (M: F= 1.3:1) and most of the children were in the age group of 6-10 years.
2. Neck was the commonest site of acanthosis nigricans.
3. BMI assessment of children with acanthosis nigricans suggested majority were overweight and obese (76%).
4. Majority of children with AN had central obesity (51%).
5. Grade 4 of neck severity and grade 3 of neck texture of children with AN were significantly associated with insulin resistance.
6. Acanthosis nigricans was associated with insulin resistance (HOMA2-IR) in normal, overweight and obese children.
7. Hence we would like to propose that all the children with acanthosis nigricans must be screened for Insulin resistance irrespective of body mass index. With this evidence of Insulin resistance, guidelines can be given to the parents about life style modification which will help in preventing the onset of diabetes mellitus later and thereby, improving the quality of life of the child.

6. Conflicts of Interest.

None.

7. Source of Funding

None.

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Cite this article: Yadav PY, Swaroop MR, Mittamedi NR, Obeidullah SM, Kalra TK, Devendra SS. Clinicoepidemiological study of acanthosis nigricans in pre and primary school children and its association with body mass index, waist circumference and insulin resistance. *IP Indian J Clin Exp Dermatol* 2022;8(3):152-161.