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ORIGINAL ARTICLE

Lipoprotein(a) identifies cardiovascular risk in childhood: The Australian Aboriginal Birth Cohort Study

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Aim: To describe the lipoprotein(a) (Lp(a))profile and its relationship to cardiovascular risk factors in Australian Aboriginal children.
Methods: A cross-sectional study within a longitudinal birth cohort study in the Darwin Health Region (Northern Territory, Australia). Subjects were Aboriginal children born between 1987 and 1990 who were re-examined between 1998 and 2001. Outcome measures were cholesterol, triglycerides, high-density lipoprotein cholesterol, low-density lipoprotein cholesterol, apoB, apoA1, apoA1/B ratio, anthropometric measures, cardiovascular disease (CVD) risk factors, maternal smoking and nutrition.
Results: At a mean age of 11.4 years, results showed that high concentrations of Lp(a) were significantly related to well-known lipid-based CVD risk factors for both boys and girls, and that only one anthropometric factor, height, was significant for girls. Non-genetic factors and maternal smoking were not found to be significant contributors to Lp(a) concentrations.
Conclusions: Lp(a) should be considered as a more effective marker of CVD than anthropometric measures, and children from families with a history of premature CVD should be regularly screened for this factor.

Key words: Aboriginal; cardiovascular risk factor; children; indigenous; lipoprotein(a).

What is already known on this topic

1 Cardiovascular disease (CVD) is a major cause of death in Australian indigenous populations.
2 CVD begins in childhood, and early risk identification is essential for effective intervention.
3 Previous research has focused on adults and has examined the relationship between anthropometric characteristics and CVD risk factors in order to determine markers of premature CVD.

What this paper adds

1 Conventional anthropometric markers were not good CVD risk markers for the Aboriginal Birth Cohort participants.
2 Serum lipoprotein(a) appears to be a more sensitive early marker of CVD than anthropometric measurement
3 Children with a family history of premature CVD should be regularly screened for serum lipoprotein(a).

Over the past three decades, the incidence of cardiovascular disease (CVD) in the indigenous (Aboriginal) population of the Northern Territory of Australia has increased and has continued to remain high during recent years.[1] These high rates of CVD have significantly reduced the average life-span of Aboriginal Territorians when compared with the rest of the Northern Territory population.[2]

The serum lipoprotein(a) (Lp(a)) has been found to influence the occurrence of CVD in a number of populations, and many studies have found that it is an independent risk factor for cerebrovascular and atherosclerotic diseases.[3–6] This fact may be associated with the finding that 'structurally, Lp(a) closely resembles low-density Lipoprotein (LDL), a well established atherogenic factor for coronary heart disease (CHD)'.[7] Research into the relationship between Lp(a) and CVD has shown that there is an increased risk of artherosclerosis when levels of Lp(a) are over 300 mg/L,[8] and in the UK, Lp(a) has been found to be a strong contributor to coronary heart disease especially for patients with higher cholesterol.[5]

In relation to indigenous studies, research examining Lp(a) concentrations in a group of Australian Aboriginals found that they were highly skewed towards lower risk levels and were on average lower than those of Australian non-indigenous subjects.[8,9] The study concluded that although Lp(a) concentrations were mainly genetically determined, other non-genetic factors, such as cigarette smoking, contributed to these concentrations.

Lp(a) has not previously been studied in Aboriginal children, and it is considered that the Aboriginal Birth Cohort (ABC) study provides an important opportunity to examine whether this CVD risk factor may be contributing to the high incidence of CVD in the Northern Territory indigenous population and whether the identification of predictors of high Lp(a) in childhood may aid in early intervention and primary prevention strategies.

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This paper hypothesises that:

1 Higher levels of Lp(a) concentrations will be related to anthropometric factors, such as higher waist size, heavier weight and higher body mass index (BMI).
2 The presence of lipid-based CVD risk factors will be significantly related to elevated levels of Lp(a).
3 Non-genetic factors such as maternal smoking and nutrition would be linked with higher levels of Lp(a).
4 The presence of infection would significantly impact on levels of Lp(a).

This paper therefore examines the relationship among Lp(a) concentrations; cardiovascular, anthropometric and clinical factors; and maternal characteristics for a cohort of indigenous children in the Northern Territory of Australia with the aims of determining the concentrations of Lp(a) in this cohort, what factors appear to be influencing those concentrations and the implications of these findings for the cohort as they get older and, ultimately, for the wider indigenous population of the Northern Territory.

Method

The recruitment and follow-up of this birth cohort has been previously published in detail.[10] In brief, 686 out of 1238 Aboriginal children born at the Royal Darwin Hospital between January 1987 and March 1990 were recruited into a cohort. There were no significant differences in the mean birthweight, birthweight frequencies or sex ratio between those recruited and not recruited.[10] The Royal Darwin Hospital is the designated hospital for normal newborn delivery within the Darwin Health Region. This study is restricted to children from the Darwin Health Region (570 of the 686 children in the original cohort).

Cohort participants were re-examined between December 1998 and March 2001. Each child was assessed while wearing light clothing and barefoot. Height was measured to the nearest millimetre with a portable stadiometer. Weight was measured to the nearest 0.1 kg with a digital electronic scale (Tanita model TBF-521, Tanita Corporation, Arlington Heights, IL, USA). Sitting blood pressure was measured using an automatic unit using an appropriate cuff size (Lifesigns BP Monitor, Welch Allyn, Sydney, NSW, Australia). Mid-upper arm and waist circumferences were measured to the nearest millimetre using a flexible tape. Waist circumference was measured at the midpoint between the lowest rib and iliac crest along the midaxillary line at the end of expiration. Triceps and sub-scapular skin folds were measured using Harpenden calipers (West Sussex, UK). These measurements were repeated in triplicate and averaged. Body composition (fat %) was measured using bioelectrical impedance analysis (TBF-310GS Body Composition Analyzer, Tanita Corporation). Onset of puberty was determined by physical examination. Children were asked to fast from midnight, and blood samples were taken in the morning after application of a topical anaesthetic cream (EMLA, Sydney, NSW, Australia). Breakfast was provided following the study visit.

Lp(a) concentrations were measured using a Beckman image as milligram per litre, and those values below the sensitivity of the assay were coded as zero in accordance with previous research.[8] Plasma glucose concentration, triglycerides and high-density lipoprotein cholesterol (HDL-c) were measured by a routine enzymatic method using (model 917, Hitachi, Tokyo, Japan) an auto-analyser using Roche reagents (Roche Diagnostics, Basel, Switzerland) at the Western Diagnostic Pathology, Darwin, Australia. Low-density lipoprotein cholesterol (LDL-c) was calculated from Friedwald's equation: LDL-c = total cholesterol – (HDL-c – (triglycerides/2.2)). Plasma insulin was measured by a two-site AIA-PACK immuno-enzymometric assay using a Tosoh AIA-600 immuno-analyser (Tosoh, Tokyo, Japan) with no cross-reactivity with pro-insulin at the Royal Perth Hospital Laboratory, Perth, Australia. Insulin resistance was estimated using the homeostasis model assessment of insulin resistance.

The anthropometric measures studied were birthweight (grams), weight (kilogram), height (centimetres), waist *z*-score, waist/height ratio, sub-scapular skin fold and triceps skin fold. The cardiovascular and metabolic risk factors were systolic and diastolic blood pressure, BMI, waist circumference and total body fat (%), apoA1 and apoB and their ratio, HDL-c, LDL-c, Lp(a) and insulin. Surrogate measures of nutrition were red cell folate and haemoglobin concentrations, and indicators of infection were the white cell count, erythrocyte sedimentation rate and the presence of sores or scabies.

Maternal smoking was measured by whether or not the mother had smoked during pregnancy. Maternal Aboriginal parentage was determined by whether the child's mother was Aboriginal.

The Joint Institutional Ethics Committee of the Royal Darwin Hospital and the Menzies School of Health Research, including the Aboriginal Ethical Sub-Committee, approved the study. Written consent was obtained from the care givers of all children.

Statistical Analysis

Statistical analysis was undertaken using the STATA software package (TX, USA). Lp(a) distributions were highly skewed towards lower levels. Spearman correlation analysis was used as a non-parametric test (significance level set at 5%) to examine the direction and strength of relationships between variables.

In order to compare the levels of Lp(a) in relation to risk factors, *t*-tests were run comparing the lowest, safest levels of Lp(a) with the highest, least safe levels by gender. The accepted convention for the cut-off of risk levels was used, with low risk at 100 mg/L or less, and high risk at 300 mg/L or more.[8]

Results

The demographic and anthropometric characteristics of the ABC children are shown in Table 1. The mean age of the cohort was 11.4 years with a mean weight of 35.8 kg and height of 143.8 cm. Not unexpectedly, girls had a significantly lower mean birthweight than boys ($P < 0.001$), but they also had a significantly higher waist *z*-score ($P = 0.006$), total body fat ($P = 0.002$), sub-scapular skin fold ($P < 0.001$) and triceps skin fold ($P = 0.01$).

There were significant differences between boys and girls for diastolic blood pressure ($P = 0.04$), homeostasis model assessment of insulin resistance ($P = 0.02$), HDL-c ($P = 0.03$), apoA1/apoB ratio ($P = 0.04$), haemoglobin ($P = 0.01$) and fasting insulin ($P = 0.01$).

Table 1 Anthropometric and demographic factors by gender

Characteristic	Total mean	Total SD	Total *n*	Boys mean	Boys SD	Boys *n*	Girls mean	Girls SD	Girls *n*	*t*-Test *P* Boys/girls
Gestational age	38.7	*1.9*	*603*	38.7	*1.9*	*306*	38.6	*1.9*	*297*	ns
Birthweight (g)	**3019.8**	**667.8**	**685**	**3110.7**	**677.3**	**354**	**2922.5**	**644.5**	**331**	**0.001**
Age	11.4	*1.13*	*506*	11.6	*1.10*	*265*	11.3	*1.15*	*241*	ns
Height	143.8	*10.53*	*505*	143.7	*10.21*	*264*	143.8	*10.88*	*241*	ns
Weight	35.8	*12.00*	*505*	35.4	*11.99*	*264*	36.3	*12.02*	*241*	ns
Waist circumference	64.6	*9.5*	*539*	64.5	*9.78*	*287*	64.7	*9.39*	*252*	ns
Waist *z*-score	**0.62**	**4.65**	**405**	**0.40**	**1.65**	**210**	**0.85**	**1.63**	**195**	**0.005**
Waist/height ratio	0.44	*0.05*	*478*	0.44	*0.05*	*251*	0.44	*0.04*	*227*	ns
Total body fat	**21.2**	**9.45**	**452**	**16.8**	**8.66**	**234**	**25.8**	**7.95**	**218**	**0.002**
BMI	16.9	*3.59*	*505*	16.7	*3.60*	*264*	17.1	*3.57*	*241*	ns
Sub-scapular skin fold	**12.0**	**8.99**	**491**	**10.7**	**9.22**	**256**	**13.4**	**8.57**	**235**	**0.001**
Triceps skin fold	**10.6**	**5.79**	**492**	**9.9**	**5.98**	**256**	**11.3**	**5.51**	**236**	**0.01**

Bold indicates significant results. BMI, body mass index; ns, not significant; SD, standard deviation.

There were only small, non-significant differences in the percentage of boys and girls with mothers who had smoked, whose mother had no Aboriginal ancestors and who had infections including sores and scabies, but there were significant differences in erythrocyte sedimentation rate ($P = 0.01$).

Birthweight was significantly positively related to Lp(a) concentrations for girls ($P = 0.05$). There were positive and significant relationships for both boys and girls between Lp(a) levels and apoB, and LDL-c and total cholesterol ($P = 0.05$). The apoA1/apoB ratio was also significant (girls $P < 0.005$, boys $P = 0.05$).

No significant difference was found in the levels of Lp(a) for boys and girls, with over half of both groups having levels of low risk and around a quarter having levels of high risk.

Anthropometric measures showed that girls with high-risk levels were significantly taller than those with low-risk levels ($P = 0.01$). Both boys and girls had significantly different LDL-c, apoB and apoA1/apoB ratio when comparing low- and high-risk categories (boys: $P < 0.001$, $P < 0.001$, $P = 0.04$; girls: $P = 0.006$, $P = 0.001$, $P = 0.004$) (Table 2).

A logistic regression showed that for boys, LDL-c was the only factor having a significant impact on low- or high-risk Lp(a) ($P < 0.001$); this was not the case for girls where none of the factors were significant. Onset of puberty was not found to be significant for boys or girls.

Discussion

This paper examined the relationship between high Lp(a) concentrations and anthropometric characteristics, and cardiovascular and metabolic risk factors and non-genetic influences. The following hypotheses were proposed:

Hypothesis 1: Higher levels of Lp(a) concentrations will be related to anthropometric factors such as higher waist size, heavier weight and higher BMI.

The findings showed that for both boys and girls, anthropometric factors such as heavier weight, higher BMI and higher waist size were not found to differ significantly for those children with higher Lp(a) concentrations with the exception of height for girls. This finding accords with other studies of non-Caucasian populations. A study of Bantu and pygmies showed no relationship among Lp(a) concentrations, age or BMI.[11,12] A Taiwanese study found that anthropometric factors were found to be not significantly associated with Lp(a) as 'lipids and Lipoprotein profiles, rather than degree of adiposity as reflected by anthropometric measures, are significantly associated with serum Lp(a) levels among school children'.[13] Generally, it has been found that 'there are no reports of the relationship between BMI or other anthropometric measures on serum Lp(a) levels'.[13]

Hypothesis 2: The presence of lipid-based CVD risk factors will be significantly related to elevated levels of Lp(a).

The current study found significant relationships between elevated levels of Lp(a) and CVD risk factors for both boys and girls, particularly LDL-c, apoB and the apoA1/apoB ratio. These findings are similar to those of Chu *et al*. (2000), who found that only lipids and lipoprotein profiles were associated with elevated Lp(a) levels and concluded that 'measurement of lipids and/or Lp(a) levels may be considered for children who have other CVD risk factors or a positive family history of premature cardiovascular disease'.[12]

The findings from the current study would therefore support regular monitoring of lipid profiles of children who have a family history of CVD.

Hypothesis 3: Non-genetic factors such as maternal smoking and nutrition would be linked with higher levels of Lp(a).

Much research has focused on the relationship of genetic factors and Lp(a), and has consequently found Lp(a) to be almost exclusively explained by genetic characteristics.[5] However, Australian studies have concluded that there are non-genetic factors that contribute to the level of Lp(a), including smoking, alcohol consumption and nutrition.[8,9] In relation to nutrition, a study of

Table 2 Lp(a) low-risk levels (≤100 mg/L) compared with high-risk levels (≥300 mg/L) by CVD risk factors, nutrition, maternal characteristics and infection by gender

	Boys (total *n* = 250)			Girls (total *n* = 221)		
	≤100 mg/L	≥300 mg/L	*P*	≤100 mg/L	≥300 mg/L	*P*
Systolic BP	108.0	106.7	ns	106.8	107.8	ns
Diastolic BP	67.6	67.9	ns	68.6	68.8	ns
Total cholesterol	4.0	4.3	ns	3.9	4.2	ns
LDL-c	**2.2**	**2.5**	**0.001**	**2.1**	**2.4**	**0.006**
HDL-c	1.2	1.2	ns	1.1	1.1	ns
Triglycerides	1.2	1.0	ns	1.2	1.2	ns
HOMA	2.0	1.5	ns	2.8	2.6	ns
ApoB	**0.70**	**0.78**	**0.001**	**0.70**	**0.78**	**0.001**
ApoA1	1.2	1.1	ns	1.1	1.1	ns
ApoB/A1 ratio	**0.58**	**0.64**	**0.04**	**0.62**	**0.72**	**0.004**
HB	128.0	128.2	ns	125.9	124.5	ns
ESR	17.9	22.1	ns	26.9	23.3	ns
White cell count	8.9	8.6	ns	8.7	9.1	ns
Fasting insulin	9.9	6.8	ns	13.2	12.4	ns
Fasting glucose	4.5	4.5	ns	4.5	4.4	ns
Folate	243.0	255.5	ns	229.1	249.2	ns
Maternal smoking	55.5	60.9	ns	54.1	49.0	ns
Maternal race	78.4	78.6	ns	83.2	86.8	ns
Sores	23.3	24.6	ns	20.6	23.1	ns
Scabies	8.2	8.7	ns	9.9	15.4	ns

Bold indicates significant results. BP, blood pressure; CVD, cardiovascular disease; ESR, erythrocyte sedimentation rate; HB, haemoglobin; HDL-c, high-density lipoprotein cholesterol; HOMA, homeostasis model assessment; LDL-c, lipoprotein cholesterol; Lp(a), lipoprotein(a); ns, not significant.

children in Turkey found that those with iron deficiency anaemia had significantly lower levels of Lp(a) than control groups.[13] The current study found no significant relationship among maternal smoking, nutrition levels or haemoglobin and Lp(a) concentrations.

Hypothesis 4: The presence of infection would significantly impact on levels of Lp(a).

There were no significant differences in Lp(a) levels for children with infection, a finding that accords with previous research that concluded that Lp(a) concentrations did not differ significantly for children with bacterial infection, although it was found to decrease apoA1 and apoB levels, and total cholesterol.[14]

Other Factors That May Influence Increased Lp(a) Concentrations

Indigenous parentage was also examined in order to determine its relationship with high Lp(a) concentrations. However, it was found that the extent of Aboriginal parentage was not significant for this group of children. Future research should include more detailed analysis of ancestry in order to better examine whether Aboriginality is a significant factor in determining Lp(a) concentrations.

Limitations of the Study

One of the limitations of the current study is that of the young age of the ABC children in that the impact of various factors on lipid concentrations may not be apparent at the age of 11 years but may become more identifiable as the children age. Further tracking of biomedical markers will be done through analysis of wave three of the study, which will compare differences between the children at 11 and 18 years of age.

Conclusion

In conclusion, the present study showed that high Lp(a) concentrations were more strongly related to well-known lipid-based CVD risk factors than to anthropometric or non-genetic factors, and that to some extent, these findings may be gender specific. It is possible that these risk factors will be more clearly identified as the children grow into their teenage years, and Lp(a) may then be found to have a stronger relationship with CVD and non-genetic factors than have been shown by the younger ABC children analysed in this paper. However, the findings suggest that Lp(a) should be considered as a more effective marker of CVD than anthropometric measures and that children from families with a history of premature CVD should be regularly screened for this factor. Given the increase in the incidence of CVD in the Northern Territory indigenous

population in recent years, further consideration should be given to introducing this course of action to help identify those children at risk of premature CVD.

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