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Relationship between metabolic control and plasma lipoprotein level in diabetic children

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All lipid and lipoprotein fractions except for total cholesterol were found to be high in diabetic children under long-term treatment. There was a positive correlation between HDL-cholesterol, HbA_{1a-c} and the insulin dose.

In untreated diabetic children, plasma triglyceride and its subfractions were also high, but phospholipids were normal. Total plasma cholesterol was normal, but its VLDL + LDL subfraction was increased and HDL-cholesterol low. After two months of insulin-treatment, parallel with the decrease of HbA_{1a-c}, the HDL-cholesterol increased and there was a drop in the level of triglycerides.

It is concluded that HDL-cholesterol is a good parameter of metabolic control during the initial therapy of diabetes. In patients under long-term treatment there is no such a simple relationship between control and HDL-cholesterol and the daily insulin-dose seems to play an important role in the regulation of plasma HDL-cholesterol.

Atherosclerotic cardiovascular disease is a leading cause of death in insulin-dependent, juvenile onset diabetics. In view of the importance of HDL-cholesterol as a negative risk factor in the development of ischaemic heart disease [7, 21], the relationship between metabolic control and serum lipids and lipoproteins has become an important aspect of diabetes.

In most surveys the incidence of fasting hyperlipoproteinaemia in diabetics has been 30 to 40% [15, 16, 19]. Beside the most frequently elevated triglyceride and cholesterol [1, 15, 16, 20, 29, 35], recently high HDL-cholesterol levels have also been described in insulin-treated maturity onset and juvenile diabetes [3, 7, 10, 17, 18, 22]. A low HDL-cholesterol level has been observed in newly diagnosed insulin deficient diabetes [31] and in maturity onset diabetes [3, 7, 17, 21]. Since HDL-cholesterol showed a significant positive correlation with the insulin sensitive lipoprotein lipase activity [22], and since HDL-cholesterol increased after one year insulin treatment in adults [25], insulin therapy was assumed to play a role in the regulation of HDL-

This work is dedicated to Professor Julius Mestyán on the occasion of this 60th birthday.

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cholesterol level in diabetics. Taskinen and Nikkilä [31] could not, however, demonstrate any increase of HDL-cholesterol level after two weeks insulin therapy in insulin deficient adult diabetics.

Insufficient control of diabetes is associated with increased lipid levels [5, 22, 28, 33]. The triglyceride and cholesterol levels showed a significant positive correlation with HbA_{1a-c} [9, 27], the fasting blood glucose level [8] and daily urinary glucose output [8] in diabetics. In a previous study [18] a similar significant positive correlation was demonstrated between HbA_{1a-c} and plasma HDL-cholesterol level in diabetic children.

The aim of the present study was to analyse the relationship between plasma lipoproteins and diabetes control, and to investigate the mechanism responsible for the elevation of HDL-cholesterol in juvenile diabetes.

Materials and Methods

Thirty two diabetic children (19 boys and 13 girls) and 48 non-diabetic children (29 boys and 19 girls) were studied. The means and ranges of ages were similar in the two groups of children, 10.1 (2–15) *vs.* 9.9 (2–15) years. Most diabetic children were followed as outpatients and some of them were admitted for checking their metabolic control. In 5 children diabetes was newly diagnosed, and in the remainder the average duration was 3.8 years (range, 1–12 years). The newly diagnosed diabetic children were treated as a separate group from those having already been treated for more than 1 year. The daily insulin dose at the time of the study varied from 0 to 1.96 U/kg. In the diabetic children the metabolic control was checked 4–5 times every year. The non-diabetic children were admitted for minor operations or with mild upper respiratory disease and blood samples taken for routine laboratory measurements were used for the purposes of this study. In diabetic children blood samples were collected after overnight fast and before the injection of the morning dose of insulin. Blood samples were taken in EDTA tubes, and plasma was separated immediately in a refrigerated centrifuge, and stored at −20 °C until estimation.

Blood glucose was estimated with the glucose oxidase method and urinary glucose was measured by the o-toluidine method. Ames Acetest tablets were used to assess urinary ketone excretion. Glycosylated haemoglobin was separated on Biorex 70 ion exchange columns. Cholesterol and triglyceride were measured enzymatically with Galenofarm and Boehringer tests. Phospholipids were measured colorimetrically [3]. HDL and VLDL + LDL were separated by heparin manganese chloride precipitation [2, 6, 7, 34]. HDL was determined in the supernatant and the combined concentration of VLDL + LDL was measured from the precipitate [2].

Results

Lipid and lipoprotein levels in diabetic children

All lipid and lipoprotein fractions except for cholesterol were significantly higher in chronic insulin treated diabetic children than in the controls (Table I).

In newly diagnosed, untreated diabetic-children plasma triglyceride and its HDL and VLDL + LDL-subfractions were significantly increased. Total cholesterol was not significantly different from controls, but VLDL +

TABLE I

	N. D. Diabetics		C. T. Diabetics		Controls	
	No.	Mean ± S. E.	No.	Mean ± S. E.	No.	Mean ± S. E.
Trigliceride, mmol/l	5	1.7±0.151[c,d]	108	1.36±0.1[b]	48	1.03±0.05
Cholesterol, mmol/l	5	4.3±0.23	122	4.1±0.14	48	3.8±0.114
Phospholipid, mmol/l	5	2.12±0.2	75	2.52±0.13[c]	48	1.72±0.09
HDL-triglyceride, mmol/l	5	0.63±0.09[c]	74	0.59±0.026[c]	48	0.38±0.02
HDL-cholesterol, mmol/l	5	0.74±0.12[c,d]	118	1.5±0.04[b]	48	1.26±0.07
HDL-phospholipid, mmol/l	5	1.12±0.15	73	1.46±0.08[c]	48	1.09±0.09
VLDL + LDL-triglyceride, mmol/l	5	0.96±0.1[b]	72	0.97±0.06[c]	48	0.72±0.045
VLDL + LDL-cholesterol, mmol'l	5	2.6±0.21[b]	72	2.37±0.12[a]	48	1.91±0.1
VLDL + LDL-phospholipid, mmol/l	5	0.9±0,1	72	0.97±0.066[a]	48	0.75±0.06

[a] $p < 0.05$, [b] $p < 0.01$, [c] $p < 0.001$ for the difference between cases and controls, [d] $p < 0.001$ for the difference between N. D. and C. T. diabetic groups. N. D.: newly diagnosed, C. T.: chronically treated

LDL-cholesterol was higher and HDL-cholesterol was much lower than in the controls. There was no change in plasma phospholipid and its HDL and VLDL + LDL subfractions (Table I).

In the untreated diabetics, high total triglyceride and low HDL-cholesterol were the two characteristic differences in the lipoprotein profiles of the two diabetic groups (Table I).

Plasma lipoprotein level and the degree of metabolic control

"Chronic" insulin treated children. Dividing the "chronic" insulin treated diabetic children into groups of satisfactory versus poor control on the basis of urinary glucose excretion, fasting blood glucose and HbA_{1a-c}, no differences in the plasma lipoproteins could be demonstrated except for the higher triglyceride level in the group with poor control (Table II).

However, when all values of the two groups were analysed, a significant positive correlation was observed between plasma cholesterol, HDL-cholesterol and glycosylated haemoglobin (Figs 1, 2). No such relationship existed between HbA_{1a-c} and triglyceride, phospholipid and their HDL- and VLDL + LDL subfractions.

Newly diagnosed diabetics. The metabolic control as judged by HbA_{1a-c}, fasting blood glucose and urinary glucose excretion, improved dramatically as a result of insulin treatment

TABLE II

Influence of satisfactory versus poor control of diabetes on plasma lipid and lipoprotein concentrations

	Satisfactory control		Poor control		
	No.	Mean ± S. E.	No.	Mean ± S. E.	p
HbA_{1a-c} per cent	51	10.2±0.5	71	18.5±0.6	<0.01
Fasting blood, glucose mmol/l	51	8.7±0.32	71	15.69±1.5	<0.01
Urinary glucose, g/day	51	12.8±2.1	71	54.0±5.4	<0.01
Urinary ketone bodies	51	neg.	71	+ − +	+++
Triglyceride, mmol/l	45	1.076±0.08	63	1.56±0.15	<0.02
Cholesterol, mml/ol	51	3.9±0.19	71	4.34±0.17	ns
Phospholipid, mmol/l	34	2.61±0.14	41	2.45±0.17	ns
HDL-triglyceride, mmol/l	33	0.57±0.04	41	0.6±0.04	ns
HDL-cholesterol, mmol/l	53	1.48±0.06	65	1.52±0.05	ns
HDL-phospholipid, mmol/l	35	1.35±0.1	38	1.55±0.12	ns
VLDL + LDL-trigliceride, mmol/l	33	0.89±0.08	39	1.02±0.08	ns
VLDL + LDL-cholesterol, mmol/l	33	2.43±0.18	39	2.31±0.15	ns
VLDL + LDL-phospholipid, mmol/l	33	0.94±0.07	39	0.99±0.097	ns

TABLE III

Effect of insulin on metabolic control and lipoprotein levels in newly diagnosed diabetics

	Before insulin		After insulin*		
	No.	Mean ± S. E.	No.	Mean ± S. E.	p
HbA_{1a-c}, per cent	5	14.56±0.6	5	10.02±1.02	<0.01
Fasting blood glucose, mmol/l	5	12.34±1.01	5	8.42±1.02	<0.01
Urinary glucose, g/day	5	37.1 ±5.4	5	19.4±4.1	<0.05
Triglyceride, mmol/l	5	1.7±0.15	5	1.13±0.1	<0.05
Cholesterol, mmol/l	5	4.3±0.23	5	3.89±0.2	ns
Phospholipid, mmol/l	5	2.12±0.2	5	2.2±0.23	ns
HDL-triglyceride, mmol/l	5	0.63±0.09	5	0.43±0.08	ns
HDL-cholesterol, mmol/l	5	0.74±0.12	5	1.36±0.13	<0.01
HDL-phospholipid, mmol/l	5	1.12±0.15	5	0.05±0.21	ns
VLDL + LDL-triglyceride, mmol/l	5	0.96±0.1	5	0.8±0.1	ns
VLDL + LDL-cholesterol, mmol/l	5	2.60±0.21	5	2.4±0.22	ns

* After two months of insulin therapy.

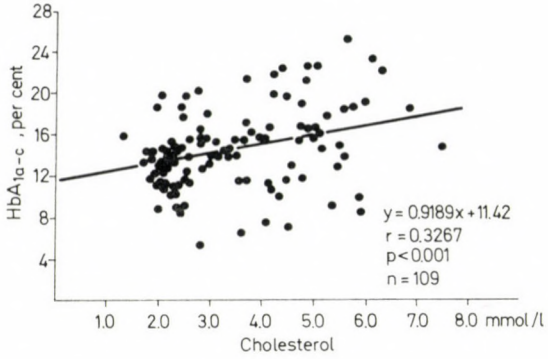

FIG. 1. Relatonship between glycosylated Hb and plasma cholesterol

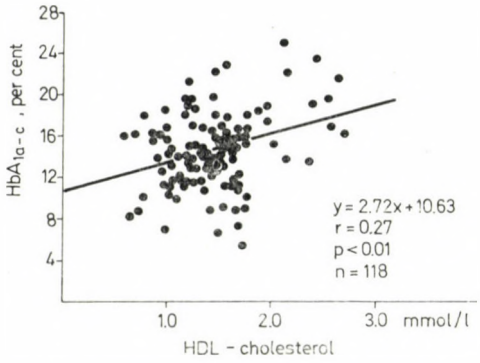

FIG. 2. Relationship between glycosylated Hb and HDL-cholesterol

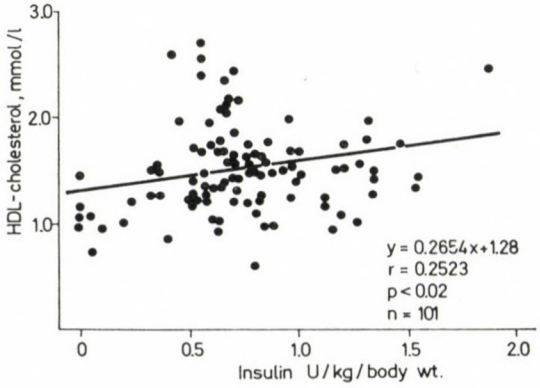

FIG. 3. Correlation between HDL-cholesterol and daily insulin dose

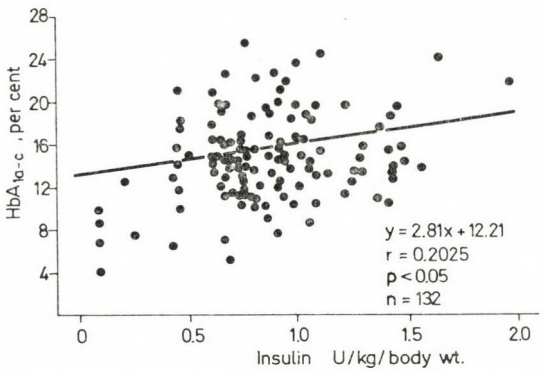

FIG. 4. Correlation between glycosylated Hb and daily insulin dose

and diet (Table III). Parallel with the improvement, HDL-cholesterol increased significantly and triglyceride decreased. The other lipids and lipoprotein subfractions did not change significantly (Table III).

In addition to the relationship between metabolic control and lipoprotein levels, the possible correlation between insulin therapy and lipoprotein levels has also been investigated.

Daily insulin dose, glycosylated Hb and plasma lipoprotein levels

The daily insulin dose showed a significant positive correlation with HDL-cholesterol (Fig. 3) in the "chronic" insulin treated diabetics suggesting that the more the insulin, the higher the HDL-cholesterol level. Higher insulin doses did not necessarily mean a better control. On the contrary, there was a positive correlation between Hba_{1a-c} and the dose of insulin (Fig. 4).

DISCUSSION

The significantly elevated lipid and lipoprotein levels observed in this study were in accordance with previous findings [8, 15, 16, 19]. There are, however, controversial data in the literature on the plasma level of HDL-cholesterol in diabetics and on its relationship with metabolic control. Significantly lower plasma HDL-cholesterol levels have been reported in maturity-onset [3, 7, 17, 21], and in untreated insulin deficient [31] diabetes. High or normal values have been observed in insulin treated maturity-onset and juvenile diabetes [3, 7, 10, 17, 22]. Elkeles et al [12] and Yudkin et al [36] found a positive, while Calvert et al [7] a negative correlation between HDL-cholesterol and glycosylated haemoglobin in diabetics. Barta et al [4] found an increase in HDL-cholesterol concentration parallel with the improvement of metabolic control, while others [31] failed to do so.

In the present study plasma HDL-cholesterol was significantly higher in "chronic" insulin treated and significantly lower in untreated diabetic children than in age-matched controls.

In addition to the fundamental difference in the plasma HDL-cholesterol levels of the two diabetic groups, further differences were revealed when the relationship between HbA_{1a-c} and HDL-cholesterol was studied. There was a positive correlation between HDL-cholesterol and HbA_{1a-c} in "chronic" insulin treated diabetic children confirming our previous observation [18]. In newly diagnosed diabetic children, however, the commencement of insulin therapy was associated with a decrease of HbA_{1a-c} and with an increase of the HDL-cholesterol level. In other words, an increased HDL-cholesterol would indicate poor control in the first and improving metabolic control in the second group. Or else, the relationship between diabetic metabolic control and the plasma HDL-cholesterol levels is a complex one and HDL-cholesterol could not be regarded as an independent parameter of the quality of control.

The significant positive correlation between HDL-cholesterol and daily insulin dose in "chronic" insulin treated children suggests that insulin therapy itself could play an important role in the regulation of plasma HDL-cholesterol level in diabetic children.

A remarkable part of HDL-cholesterol is a product of the catabolism of triglyceride-rich lipoproteins [11, 14]. Lipoprotein lipase, the main catabolic enzyme of the triglyceride rich lipoproteins is known to be highly sensitive to insulin [23, 26, 32]. Low lipoprotein lipase (LPL) activity with low HDL-cholesterol level in untreated insulin deficient diabetics [31], and high LPL activity with high HDL-cholesterol level in chronic insulin-treated diabetics [22] have been reported. LPL activity and HDL-cholesterol concentration showed a strong positive correlation [22, 31]. Thus, exogenous insulin probably elevates the HDL-cholesterol level by increasing LPL activity.

On the basis of our observations it seems likely that plasma HDL-cholesterol levels in children are influenced both by exogenous insulin as well as by other factors known to alter the plasma HDL-cholesterol concentration. On the one hand, insulin treatment was followed by a rapid improvement of metabolic control and an increase of HDL-cholesterol as shown in the newly-diagnosed group of diabetic children (Table III). High HDL-cholesterol levels, on the other hand, could also be associated with poor control despite the high dose of insulin as found in the "chronic" insulin treated group (Figs 3, 4). The opposite process, increasing HDL-cholesterol levels parallel with decreased insulin dosage has also been observed in diabetic children: Barta et al [4] observed a significant increase in plasma HDL-cholesterol during a summer camp for diabetic children when the insulin dose had been decreased.

Finally, our observations proved further data to the complexity of the independent and interlinked factors influencing plasma lipoprotein levels in diabetes. We are well aware that these observations require further confirmation in a more homogeneous group of young insulin dependent diabetics, especially in view of the conflicting findings of others.

References

1. Albrink MJ, Lavietes PH, Man, EB: Vascular disease and serum lipids in diabetes mellitus. Observations over 30 years (1931–1961). Ann Intern Med 58: 305, 1963
2. Andersen GE, Friis-Hansen B: Cord serum lipid and lipoprotein-cholesterol values in normal and beta-methasone-treated newborns of varying gestational age. Acta Paediatr Scand 66:355, 1977
3. Bar-On H, Landau D, Berry E: Serum-high-density lipoprotein and university group Diabetes Program results. Lancet 1:761, 1977
4. Barta L, Molnár M, Tichy M: Diabeteszes gyermekek táborozásának befolyása az anyagcsere-állapotra. Orv Hetil 112: 1189, 1981
5. Bennion LJ, Grundy SM: Effects of diabetes mellitus on cholesterol metabolism in men. N Engl J Med 296:1365, 1977
6. Bullock DG, Carter TJN: HDL cholesterol analysis. Lancet 1:154, 1979
7. Calvert GD, Graham JL, Mannik T, Wise PH, Yeates RA: Effects of therapy on plasma high-density-lipoprotein-cholesterol concentration in diabetes mellitus. Lancet 2:66, 1978
8. Chase HP, Glasgow AM: Juvenile diabetes mellitus and serum lipids and lipoprotein levels. Am J Dis Child 130: 1113, 1966
9. Ditzel J, Kjaergaard J: Hemoglobin A_{1c} concentrations after initial insulin treatment for newly discovered diabetes. Br Med J 1:741, 1978
10. Durrington P: HDL-cholesterol in diabetes mellitus. Lancet 2:206, 1978
11. Eisenberg S, Schurr D: Phospholipid removal during degradation of rat plasma very low density lipoprotein in vitro. J Lipid Res 16:341, 1975
12. Elkeles RS, Wu J, Hambley J: Haemoglobin A_1, blood glucose and high density lipoprotein cholesterol in insulin requiring diabetes. Lancet 2:547, 1978
13. Field JB: Extraction of insulin by the liver. Annu Rev Med 24:409, 1973
14. Havel RJ, Kane JP, Kashyap ML: Interchange of apoproteins between chylomicrons and high density lipoproteins during alimentary lipemia in man. J Clin Invest 52:32, 1973
15. Hayes TM: Plasma lipoproteins in adult diabetes. Clin Endocrinol (Oxf) 1:247, 1972
16. Kaufmann RL, Assal JPh, Soeldner JS, Wilmhurst EG, Lemaire JR, Gleason RE, White R: Plasma lipid levels in diabetic children. Effect of diet restricted in cholesterol and saturated fats. Diabetes 24:672, 1975
17. Kennedy AL, Lappin TRJ, Lavery TD, Hadden DR, Weaver JA, Montgomery DAD: Relation of high density lipoprotein cholesterol concentration to type of diabetes and its control. Br Med J 2:1191, 1978
18. Klujber L, Molnár D, Kardos M, Jászai V, Soltész G, Mestyán J: Metabolic control, glycosylated hemoglobin and high density lipoprotein cholesterol in diabetic children. Eur J Pediatr 132:289, 1979
19. Leonhardt W, Hanefeld M, Haller H, Jaross W: Implications of hyperlipoproteinemia, diabetes and obesity. Proc. 2. European Meeting of Metabolism, Padova 1977 P. 383
20. Lewis B, Mancini M, Mattock M, Chait A, Fraser TR: Plasma triglyceride and fatty acid metabolism in diabetes mellitus. Eur J Clin Invest 2:445, 1972
21. Lopes-Virella MF, Stone PG, Colwell JA: Serum high density lipoprotein in diabetic patients. Diabetologia 13:285, 1977
22. Nikkilä EA, Hormila P: Serum lipids and lipoproteins in insulin-treated diabetes. Demonstration of increased high density lipoprotein concentrations. Diabetes 27:1078, 1978
23. Nilsson-Ehle P, Carlstom S, Belfrage P: Rapid effects on lipoprotein lipase activity in adipose tissue of humans after carbohydrate and lipid intake. Time course and relation to plasma glycerol, triglyceride and insulin levels. Scand J Clin Lab Invest 35:373, 1975
24. Olefsky JM, Farquhar JW, Reaven GM: Reapraisal of the role of insulin in hypertriglyceridemia. Am J Med 57:551, 1974
25. Paisey R, Elkeles RS, Hambley J, Magill P: The effects of chlorpropamide

and insulin on serum lipids lipoproteins and fractional triglyceride removal. Diabetologia 15:81, 1978
26. Persson B, Hood B, Angervall G: Effects of prolonged fast on lipoprotein lipase eluted from human adipose tissue. Acta Med Scand 188:225, 1970
27. Peterson CM, Koenig RJ, Jones RL, Saudek CD, Cerami A: Correlation of serum triglyceride levels and hemoglobin A_{1c} concentration in diabetes mellitus. Diabetes 26:507, 1977
28. Schrade W, Boehle E, Biegler R, Harmuth E: Fatty-acid composition of lipid fractions in diabetic serum. Lancet 1:285, 1963
29. Sharma D, Bansal BC, Prakash C: Serum lipid studies in thin insulin-dependent diabetics below the age of 30 years. J Indian Med Assoc 54:416, 1970
30. Steele BW, Kochler DF, Azar MM, Blaszkowsky TB, Kuba K, Dempsey ME: Enzymatic determination of cholesterol in high-density lipoprotein fractions prepared by a precipitation technique. Clin Chem 22:98, 1976
31. Taskinen MR, Nikkilä EA: Lipoprotein lipase activity of adipose tissue and skeletal muscle in insulin-deficient human diabetes. Diabetologia 17:351, 1979
32. Taskinen MR, Nikkilä EA: Effects of caloric restriction on lipid metabolism in man: Change of tissue lipoprotein lipase activities and of serum lipoproteins. Atherosclerosis 32:289, 1979
33. Traisman HS, Newcomb AL, Sever JL, Hammes R: Blood lipid and protein levels in juvenile diabetes mellitus. Diabetes 9:481, 1960
34. Van Der Haar F, Van Gent CM, Schouten FM, Van Der Voorth HA: Methods for the estimation of high density cholesterol, comparison between two laboratories. Clin Chim Acta 88: 469, 1978
35. Wahl P, Hasslacher Ch, Vollmar J: Diabetes and Hyperlipoproteinämien. Dtsch Med Wochenschr 99:2158, 1971
36. Yudkin JS, Boucher BJ, France MW, Welch SG, Swindlehurst C: The relationship between concentrations of glycosylated haemoglobins and of serum high density-lipoprotein cholesterol in diabetic patients. Clin Sci 56:269, 1979

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