# Cholesterol ester transfer protein in children on peritoneal dialysis

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# CHOLESTEROL ESTER TRANSFER PROTEIN IN CHILDREN ON PERITONEAL DIALYSIS

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♦ ***Objectives:*** **To examine whether cholesterol ester transfer protein (CETP) activity and mass contribute to dyslipidemia in children on peritoneal dialysis (PD), and to determine whether CETP activity or mass is responsible for severer hyperlipidemia in smaller (younger) patients.**

♦ ***Study Design:*** **27 patients (18 males, 9 females; mean age 11.8 ± 6.1 years) were enrolled. Each patient had been receiving PD for more than 6 months. Fasting blood samples were drawn and CETP activity, CETP mass, total cholesterol, triglyceride, β-lipoprotein profiles, lipoprotein lipid profiles (cholesterol and triglyceride in lipoproteins), apoprotein profile, and serum albumin levels were measured. The results were then compared, using Student's t-test, with those for a control group. In the patient group, the relationships between CETP activity and each factor were examined using simple and multiple regression analyses.**

♦ ***Results:*** **Total cholesterol, triglyceride, low-density lipoprotein (LDL), very low-density lipoprotein (VLDL), chylomicron, and ApoB levels were significantly higher in the patient group. Mean CETP activity levels were 106% ± 24% in the patient group and 111% ± 21% in the control group. No significant difference in CETP activity was seen between the two groups, but CETP mass was lower in the patient group than in the control group (2.2 ± 0.6 µL/dL for the patient group vs 2.8 ± 0.9 µL/dL for the control group, $p = 0.01$). As a result, specific CETP activity (activity/protein mass ratio) was significantly higher in the patient group ($p < 0.0001$). CETP activity was positively related to LDL and other atherogenic factors and negatively related to serum albumin level. No relationship between CETP activity and patient body weight was seen.**

♦ ***Conclusion:*** **Specific CETP activity was higher in the patient group compared with that in the control group, and strong correlations were found between CETP activity and atherogenic factors in the patient group. Therefore, CETP seems to be associated with lipid abnormalities in children on PD but is not responsible for the severer hyperlipidemia seen in smaller children.**

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KEY WORDS: Cholesterol ester transfer protein (CETP); specific CETP activity; children; hyperlipidemia; serum albumin.

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Japanese children on peritoneal dialysis (PD) require longer periods of dialysis than patients in western countries because they have a lower chance of receiving a cadaver transplant. As a result, hyperlipidemia as a complication of PD cannot be ignored, even in children.

Several studies have described the causes of hyperlipidemia in patients receiving PD: (a) a decreased lipoprotein catabolic rate due to decreased lipoprotein lipase activity (1), (b) glucose absorption from dialysate (2,3), (c) an increased lipid production rate secondary to protein loss via dialysate or hypoalbuminemia (3–5).

In our previous study (6), smaller (generally younger) patients exhibited severer hyperlipidemia and their cholesterol and triglyceride levels were much higher than those of adult patients, suggesting that some specific etiology must be responsible for childhood hyperlipidemia in PD patients. The reasons for the negative correlations between body weight and serum cholesterol and triglyceride levels in children on PD are not clearly understood. Our previous study suggested that it was not protein loss via dialysate or serum albumin levels, but that their energy intake per body weight strongly influenced hypertriglyceridemia (6). However, we were unable to identify any definite factors contributing to hypercholesterolemia.

Matsuyama *et al.* described the differences in cholesterol and triglyceride composition in lipoproteins between normal control children and pediatric patients receiving PD (7). The cholesterol and triglyceride contents of triglyceride-rich lipoprotein, intermediate-density lipoprotein, and low-density lipoprotein (LDL) were much higher, but the cholesterol content of high-density lipoprotein 2 (HDL2) was significantly lower in the patient group than in the control group.

Since cholesterol ester transfer protein (CETP) transfers cholesterol ester from HDL to apolipoprotein B (ApoB)-containing lipoproteins, reducing HDL, CETP is likely to contribute to the hyperlipidemia seen in children on PD. CETP is an important determinant of lipoprotein composition because it mediates the net transfer of triglycerides and cholesterol ester between lipoproteins. CETP plays an important role in reverse cholesterol transport by transferring cholesterol ester from HDL to ApoB-containing lipoproteins such as LDL and very low-density lipoprotein (VLDL). In this light, the role of CETP can be said to be "antiatherogenic." On the other hand, LDL and VLDL are considered proatherogenic; by increasing the levels of LDL and VLDL, CETP also seems to play an "atherogenic" role. Whether CETP should be classified as an antiatherogenic or an atherogenic protein remains controversial.

Asayama *et al.* concluded that CETP activity in children on continuous ambulatory peritoneal dialysis (CAPD) was excessive and it made the children's lipid environment more atherogenic (8). In their discussion, they suggested that the increased CETP activity in children on CAPD might be secondarily caused by the hyperlipidemia itself, or by overproduction of protein in the liver as a result of protein loss via the dialysate.

The aim of this study was to determine the implications of CETP activity (which expresses the net transfer capacity of cholesterol esters between lipoproteins) and CETP mass (the concentration of protein in the serum) in the hyperlipidemia of PD patients. Furthermore, the effects of these factors on the severer hyperlipidemia in younger children were also examined.

## PATIENTS AND METHODS

Twenty-seven patients (18 males, 9 females) who had been followed at the Tokyo Metropolitan Kiyose Children's Hospital were enrolled in the study. The number of patients was judged sufficient because this number was equivalent to about 15% of all pediatric patients receiving PD in Japan. When examined using a partial correlation coefficient, significant differences between the patient and control groups were observed for some factors, even with this sample size. For example, the total cholesterol level exhibited a difference at $n = 14$ ($\alpha = 0.05$, $\beta = 0.2$).

The mean age of the patients was 11.8 ± 6.1 years (2.3 – 22.4 years) and each had been on PD for more than 6 months. All the patients were anuric at the time of examination. Their original diseases consisted of focal segmental glomerular sclerosis in 7 patients, hypoplastic or dysplastic kidney in 5 patients, rapid progressive glomerular sclerosis in 4 patients, congenital nephrotic syndrome in 4 patients, hemolytic uremic syndrome in 2 patients, nephronophthisis in 2 patients, and other in 4 patients. None of the patients were taking lipid-lowering agents, steroids, or other drugs that can alter lipid metabolism, and none of them had experienced any episodes of peritonitis or surgical operations within the preceding 3 months. The method of dialysis was decided on an individual basis according to the blood pressure, desired ultrafiltration volume, peritoneal function, and lifestyle of each patient. The concentration of the dialysate required to produce the desired dry weight was determined each day according to the patient's morning body weight and blood pressure. Eight patients were on CAPD and 19 patients were on automated PD. CAPD was used mainly for infants, with a dose of 50 mL/kg of dialysate changed four or five times per day. Automated PD was performed as continuous cycling PD (CCPD) in 14 patients and 5 patients were on nocturnal PD. Five to six cycles of exchanges with 50 mL/kg/cycle, or 1100 – 1200 mL/$m^2$/cycle, of dialysate were performed in 9 – 10 hours. Five CCPD patients required extra CAPD.

Three patients were receiving tube feeding. None of the patients were receiving a special diet.

The control group (15 males, 10 females; mean age 11.5 ± 6.1 years) consisted of 25 children and student volunteers who did not have any diseases that might affect lipid metabolism. Informed consent was obtained from each control subject or from their parents. The mean age of the control group was similar to that of the patient group. Although the age matching between the two groups was not strictly 1:1, the age distributions of the two groups were nearly the same. The control subjects had an ordinary diet and their body weight and height were around average for their age.

After more than 8 hours of fasting, blood samples were drawn from both the patient and the control groups, and the serum CETP mass, CETP activity, total cholesterol, triglyceride, β-lipoprotein profiles (LDL, VLDL, chylomicron), lipoprotein lipid profiles (cholesterol and triglyceride in lipoproteins), apoprotein profiles, and serum albumin levels were measured.

The CETP activity was measured using the 14C-cholesterol ester transfer method (9), and CETP mass was measured using the enzyme-linked immunosorbent assay (ELISA) method (10) at Biomedical Laboratories, Inc. (Tokyo, Japan).

Serum total cholesterol, triglyceride, and albumin levels were measured with a Toshiba FCR automatic analyzer (Tokyo, Japan). The β-lipoprotein profile and the lipoprotein lipid profile were measured by gradient ultracentrifugation; the apoprotein levels were measured by turbidimetric immunoassay at Mitsubishi Kagaku Bio-Clinical Laboratories, Inc. (Tokyo, Japan).

STATISTICAL ANALYSIS

The differences between the control group and the patient group were quantified using the Student's t-test, and the relationships between CETP and the factors were quantified using simple regression analysis and multiple linear regression analysis. All calculations were performed using StatView 5.0J (SAS, Cary, North Carolina, USA.)

## RESULTS

The total cholesterol, triglyceride, lipoprotein profiles, and apoprotein levels of the patient group and control group are shown in Table 1. The serum albumin levels were lower in the patient group than the control group. Atherogenic factors, such as total cholesterol, triglyceride, LDL, VLDL, chylomicron, and ApoB were significantly higher in the patient group than in the control group. The triglyceride and cholesterol contents of LDL and VLDL were also much higher in the patient group than in the control group (Table 2). As Figure 1 shows, a negative correlation was seen between body weight and total cholesterol as well as triglyceride levels in the patient group but not in the control group.

The mean level of CETP activity was 106% ± 24% in the patient group and 111% ± 21% in the control group. No differences in CETP activity were seen between the two groups, but CETP mass was lower in the patient group than in the control group (2.2 ± 0.6 µL/dL for the patient group vs 2.8 ± 0.9 µL/dL for the control group, $p$ = 0.01). Figure 2 shows that specific CETP activity (activity/protein mass ratio) was significantly higher in the patient group.

TABLE 1
Lipids, Serum Albumin, and Apoproteins in Pediatric CAPD Patient Group and Control Group

| | Patients (mg/dL) | Control (mg/dL) | $p$ Value |
|---|---|---|---|
| Total cholesterol | 214±45 | 170±34 | 0.0004 |
| Triglyceride | 123±57 | 57±31 | <0.0001 |
| Serum albumin | 3.7±0.3 | 4.6±0.3 | <0.0001 |
| $ApoA_1$ | 140±20 | 145±21 | NS |
| ApoB | 97±22 | 75±15 | 0.001 |
| ApoC2/C3 | 0.33±0.1 | 0.40±0.2 | NS |
| ApoE | 4.5±1.2 | 3.9±1.0 | NS |

Apo = apolipoprotein; NS = not significant.

Figure 3 shows the relationship between specific CETP activity and LDL. A strong negative correlation was seen in the patient group. Specific CETP

TABLE 2
Lipoprotein Profiles in Pediatric CAPD Patient Group and Control Group

| | Patients (mg/dL) | Control (mg/dL) | $p$ Value |
|---|---|---|---|
| HDL-C | 41.8±10.2 | 44.7±10.0 | NS |
| HDL-TG | 9.3±5.8 | 7.8±8.2 | NS |
| LDL | 424±140 | 272±49 | <0.0001 |
| LDL-C | 106.5±49.0 | 85.0±26.2 | 0.06 |
| LDL-TG | 32.5±32.3 | 18.4±15.9 | 0.05 |
| VLDL | 188±83 | 133±49 | 0.006 |
| VLDL-C | 35.5±19.3 | 23.6±7.0 | 0.001 |
| VLDL-TG | 188.0±84.5 | 30.8±5.1 | 0.0002 |
| Chylomicron | 134.0±112.0 | 56.9±42.0 | 0.002 |

HDL = high-density lipoprotein; C = cholesterol in lipoprotein; TG = triglyceride in lipoprotein; LDL = low-density lipoprotein; VLDL = very low-density lipoprotein.

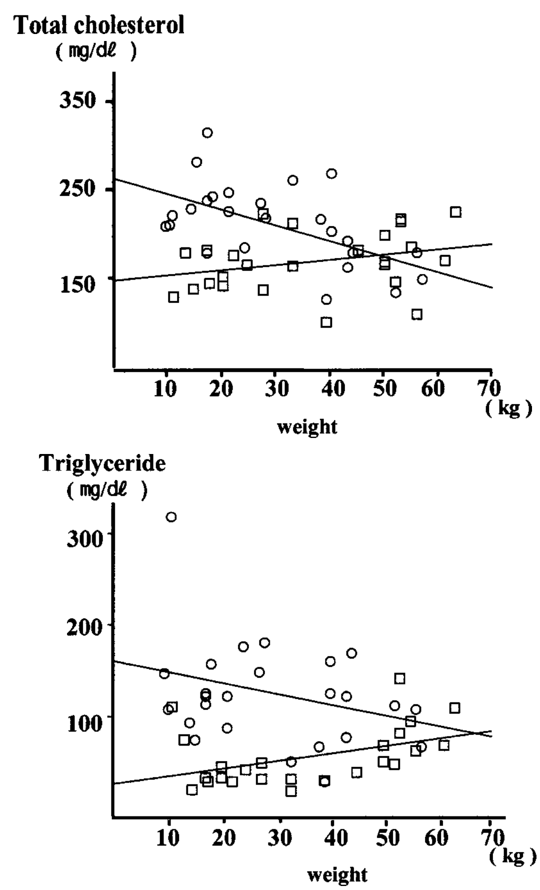


Figure 1 — Serum total cholesterol and triglyceride levels showed negative correlation with body weight in pediatric patients (circles) on CAPD (cholesterol $r$ = –0.57, $p$ = 0.003; triglyceride $r$ = –0.32, $p$ =0.12), and positive correlation in the control group (squares) (cholesterol $r$ = 0.30, $p$ = 0.15; triglyceride $r$ = 0.44, $p$ = 0.03).

activity in the patient group was also negatively correlated with some of the other atherogenic factors, such as total cholesterol, ApoB, LDL-cholesterol, and VLDL-cholesterol (data not shown). These relationships were not seen in the control group.

To assess the role of CETP in children on PD, a single regression analysis was performed to test for correlations between CETP activity (not specific activity) and lipoprotein profiles, apoproteins, and serum albumin levels in each group (Table 3). Strong correlations were found between CETP activity and the atherogenic factors total cholesterol, LDL-cholesterol, and ApoB in the patient group. On the other hand, the factors that showed significant correlation with CETP activity were HDL-cholesterol, and ApoA. No statistical significance between CETP activity and triglyceride, VLDL, C2/C3 ratio, weight, or serum albumin levels was seen in either group. We also examined the relations between CETP mass and each of the factors (data not shown), with almost the same results.

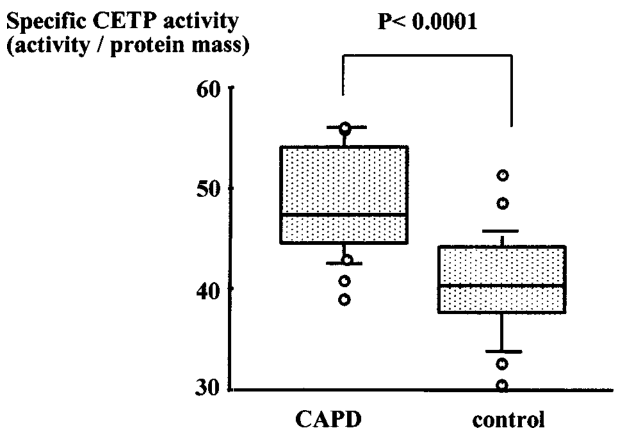


Figure 2 — Difference in cholesterol ester transfer protein (CETP) specific activity between pediatric patients on CAPD and the control group.

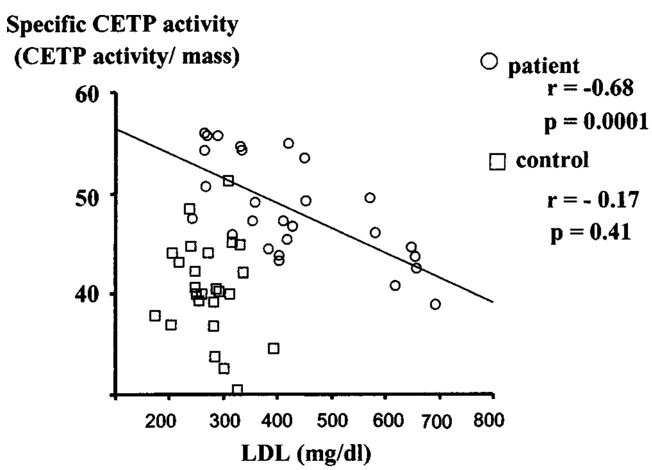


Figure 3 — Relationship between specific cholesterol ester transfer protein (CETP) activity and low-density lipoprotein (LDL) in pediatric patients on CAPD.

Table 4 shows the results of the multiple linear regression analysis for CETP activity (not specific activity) versus each factor in the patient and control groups. A forward elimination procedure of stepwise analysis was used. Total cholesterol, triglyceride, LDL, VLDL, HDL-cholesterol, serum albumin, and weight were treated as independent valuables. The most strongly correlated factor was LDL in the patient group. Although albumin level was not statistically correlated with CETP activity in the single regression analysis, a significant negative correlation was seen in the multiple regression analysis. On the other hand, HDL-cholesterol was the only affective factor in the control group. Weight was not correlated with CETP activity in either group.

TABLE 3
Correlations Between Cholesterol Ester Transfer Protein Activity and Factors in Pediatric CAPD Patients

| | Patients | | Control | |
|---|---|---|---|---|
| | *r* | *p* Value | *r* | *p* Value |
| Total cholesterol | 0.51 | 0.009 | 0.42 | 0.04 |
| Triglyceride | 0.06 | — | 0.30 | — |
| LDL | 0.67 | 0.0001 | 0.36 | — |
| VLDL | –0.14 | — | 0.01 | — |
| HDL-C | 0.001 | — | 0.60 | 0.002 |
| HDL-C/LDL-C | –0.54 | 0.003 | 0.28 | — |
| $ApoA_1$ | –0.062 | — | 0.53 | 0.007 |
| ApoB | 0.58 | 0.003 | 0.26 | — |
| ApoC2/C3 | –0.20 | — | 0.16 | — |
| ApoE | 0.48 | 0.012 | 0.19 | — |
| Weight | –0.37 | — | –0.20 | — |
| Serum albumin | –0.22 | — | 0.04 | — |

LDL = low-density lipoprotein; VLDL = very low-density lipoprotein; HDL = high-density lipoprotein; C = cholesterol in lipoprotein; Apo = apolipoprotein.

TABLE 4
Standardized Partial Regression Coefficients of Multiple Regression Analysis with the Forward Elimination Procedure, Stepwise Analysis

| Independent variable | Patients (*n*=24) | *p* Value | Control (*n*=25) | *p* Value |
|---|---|---|---|---|
| LDL | 0.69 | <0.001 | — | |
| Albumin | –0.32 | <0.05 | — | |
| HDL-C | — | | 0.56 | <0.005 |
| $R^2$ | 0.52 | | 0.35 | |

$R^2$ = determination coefficient.
Dependent variable was cholesterol ester transfer protein activity; independent variables were LDL, VLDL, HDL-C, total cholesterol, triglyceride, serum albumin, and body weight.

## DISCUSSION

In the present study, CETP activity in children receiving PD was not elevated and the CETP mass was lower than in the control group. The specific activity of CETP (activity/protein mass ratio) was significantly higher in the patient group. The factors that had the greatest impact on CETP activity and mass in the patient group were atherogenic factors, such as LDL. These results suggest that CETP is a contributing but not a major cause of lipid abnormalities in PD patients. CETP did not explain the high total cholesterol and triglyceride levels found in younger patients, which was our major concern.

As in previous reports (6,7), serum total cholesterol and triglyceride levels were higher in the patient group than in the control group, especially in smaller children. Apoprotein B, β-lipoproteins, and the cholesterol and triglyceride contents of β-lipoprotein were significantly higher in the patient group than in the control group; HDL-cholesterol levels were lower. The serum lipid profile in the patients could be described as an "atherogenic environment."

Even in atherogenic environments, however, CETP activity level itself did not differ from that in the control group. This finding was unexpected and could not be explained. However, CETP activity was strongly correlated with atherogenic factors such as LDL and ApoB. CETP seems to be one of several possible factors that contribute to the lipid abnormalities seen in pediatric patients receiving PD.

On the other hand, in the control group, HDL exhibited the strongest correlation with CETP activity. This result can be explained by the fact that, in the normal lipid environment, CETP performs reverse transport of cholesterol ester with HDL.

A large variety of specific factors can modulate CETP. To assess the role of CETP more accurately in various lipid environments, Lagrost suggested that CETP should be measured using two different approaches (10): by ELISA, which reflects the quantity of protein, and by activity, which reflects cholesterol ester transport capacity. As far as we know, CETP mass has not been previously measured in patients receiving PD. In the present study, CETP mass was lower in the patient group than in the control group. As described above, our data did not show an elevation of CETP activity in the patient group. Interestingly, however, specific activity (activity/protein mass ratio) was significantly higher in the patient group than in the control group.

Asayama *et al.* speculated that the elevation of CETP activity might be affected by accelerated protein production as a result of protein leakage via the dialysate (8). The results of the present study refute Asayama's hypothesis because the protein mass was lower in the patient group than in the control group; however, multiple regression analysis showed a negative correlation between CETP activity and serum albumin levels in the patient group. These findings suggest the existence of some form of association between hypoalbuminemia or protein loss and CETP activity. The reason why albumin showed significance in multiple regression analysis but not in simple regression analysis can be explained by the difference between the simple correlation coefficient and the partial correlation coefficient. In hypoalbuminemic diseases, such as nephrotic syndrome and congenital analbuminemia, CETP activity is negatively correlated with serum albumin level (11,12). In previous reports, hypoalbuminemia was shown to increase the nonesterified fatty acids (NEFA) that bind to lipoproteins and, because the lipoprotein surface is negatively charged, interaction with CETP easily occurs. As a result, the transport of cholesterol ester to LDL or other lipoproteins increases. In children on PD, similar lipoprotein changes resulting from hypoalbuminemia might accelerate CETP activity; however, this theory remains speculative because NEFA has not been studied in children receiving PD.

As shown in Figure 3, specific CETP activity negatively correlated with LDL and other atherogenic factors in the patient group. These findings were not observed in the control group, in which the lipid environment was normal. Essentially, CETP carries cholesterol ester from HDL to atherogenic lipoproteins, which is disadvantageous to the human body if excessive lipids are present. CETP activity is considered rate limiting *in vivo* (13). Specific activity decreased as atherogenic factor increased in the patient group, suggesting an inhibitory physiological mechanism. Mendez *et al.* demonstrated the presence of a CETP inhibitor in hemodialysis patients (14). Such an inhibitor might also be present in patients receiving PD, although no reports of an inhibitor in PD patients have been made. Morton reported that lipoprotein transfer inhibitor protein (LTIP) is strongly associated with LDL (15). The strong inverse correlation between specific CETP and LDL in our result could be explained by LTIP, if such an inhibitor were to be found in patients receiving PD.

In the present study, strong correlations were found between CETP activity and atherogenic factors in pediatric patients receiving PD. This report is the first description of the simultaneous measurement of CETP activity and CETP mass in patients receiving PD. We found that CETP activity in the patient group did not differ from that in the control group; CETP mass was lower in the patient group than in the control group, and the activity/protein mass ratio was significantly higher. CETP activity was strongly correlated with some atherogenic factors in the patient group; therefore, CETP seems to be associated with the lipid abnormalities seen in children on PD. The lipid abnormalities could not be explained, however, on the basis of CETP alone. CETP could also not explain the severer hyperlipidemia seen in younger patients. Further investigations on such topics as the synthesis of cholesterol should be made.

## REFERENCES


1. Shoji T, Nishizawa Y, Nishitani H, Yamakawa M, Morii H. Roles of hypoalbuminemia and lipoprotein lipase on hyperlipoproteinemia in continuous ambulatory peritoneal dialysis. *Metabolism* 1991; 40:1202–8.
2. Kaysen GA. Hyperlipidemia of chronic renal failure. *Blood Purif* 1994; 12:60–7.
3. Heimburger O, Stenvinkel P, Berglund L, Tranaeus A, Lindholm B. Increased plasma lipoprotein(a) in continuous ambulatory peritoneal dialysis is related to peritoneal transport of proteins and glucose. *Nephron* 1996; 72:135–44.
4. Broyer M, Niaudet P, Champion G, Jean G, Chopin N, Czernichow P. Nutritional and metabolic studies in children on continuous ambulatory peritoneal dialysis. *Kidney Int Suppl* 1983; 15:S106–10.
5. Scolnik D, Balfe JW. Initial hypoalbuminemia and hyperlipidemia persist during chronic peritoneal dialysis in children. *Perit Dial Int* 1993; 13:136–9.
6. Tanaka Y, Hataya H, Aaki Y, Ikeda M, Matsuyama T, Honda M. Hyperlipidemia in children on peritoneal dialysis: effect of energy intake on serum triglyceride. *Perit Dial Int* 2002; 22:426–8.
7. Matsuyama T, Honda M, Ito H. Lipoprotein and apoprotein abnormalities in children undergoing CAPD. In: Matsuyama T, Nakahara C, Honda M, Kamiyama Y, Hasegawa O, Ito H, *et al.*, eds. Current Concepts in Peritoneal Dialysis. Tokyo: Excerpta Medica; 1992: 555–9.
8. Asayama K, Hayashibe H, Mishiku Y, Honda M, Ito H, Nakazawa S. Increased activity of plasma cholesteryl ester transfer protein in children with end-stage renal disease receiving continuous ambulatory peritoneal dialysis. *Nephron* 1996; 72:231–6.
9. Albers JJ, Tollefson JH, Chen CH, Steinmetz A. Isolation and characterization of human plasma lipid transfer proteins. *Arteriosclerosis* 1984; 4:49–58.
10. Lagrost L. Determination of the mass concentration and activity of the plasma cholesteryl ester transfer protein (CETP). *Methods Mol Biol* 1998; 110:231–41.
11. Braschi S, Masson D, Rostoker G, Florentin E, Athias A, Martin C, *et al*. Role of lipoprotein-bound NEFAs in enhancing the specific activity of plasma CETP in the nephrotic syndrome. *Arterioscler Thromb* 1997; 17: 2559–67.
12. Braschi S, Lagrost L, Florentin E, Martein C, Athias A, Gambert P, *et al*. Increased cholesteryl ester transfer activity in plasma from analbuminemic patients. *Arterioscler Thromb Vasc Biol* 1996; 16:441–9.
13. Barter P. CETP and atherosclerosis. *Arterioscler Thromb Vasc Biol* 2000; 20:2029–31.
14. Mendez AJ, Perez GO, Hsia SL. Defect in cholesteryl ester transport in serum of patients with uremia receiving maintenance hemodialysis: increased inhibitor activity for cholesteryl ester transfer. *J Lab Clin Med* 1988; 111:677–83.
15. Morton RE. Cholesteryl ester transfer protein and its plasma regulator: lipid transfer inhibitor protein. *Curr Opin Lipidol* 1999; 10:321–7.