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The Effect of Vitamin B_6 Deficiency and Age on Plasma Cholesterol Profile in Intensely Exercised Rats

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Abstract

The purpose of this study was to determine whether vitamin B_6 deficiency and age affect the blood cholesterol profile in exercising rats. Fifty four rats were fed either a vitamin B_6 deficient diet($-B_6$) or a control diet($+B_6$) for 6 weeks, then subdivided into 3 groups: non-exercise group(NE), exercise and sacrifice group (ES), exercise and recuperation group(ER). ES group was exercised on treadmill(10^0, 0.5~0.8km/h) for 2 hours and sacrifice. ER group was recuperated three days with respective diet after exercise. At week 3 and 6, the level of plasma total cholesterol(TC), high density lipoprotein cholesterol(HDL-C) and low density lipoprotein cholesterol(LDL-C) were compared. In NE group, there was no difference in the levels of TC, HDL-C and LDL-C between $+B_6$ rats and $-B_6$ rats. The plasma levels of TC and LDL-C of 6 weeks were higher than those of 3 weeks and no difference in HDL-C between 3 weeks rats and 6 weeks rats. In ES group, there was also no difference in the levels of TC, HDL-C and LDL-C between $+B_6$ rats and $-B_6$ rats and there was no difference in TC, LDL-cholesterol between 3 weeks rats and 6 weeks rats. The level of HDL-C of 6 weeks was lower than that of 3 weeks rats. In ER group, there was no difference in the levels of TC and LDL-C not only between $+B_6$ rats and $-B_6$ rats but also between 3 weeks rats and 6 weeks rats. The level of HDL-C was lower in $-B_6$ rats than that in $+B_6$ rats and higher in 6 weeks rats than in 3 weeks rats. These results suggest that vitamin B_6 deficiency may affect the HDL-C during exercise and after recuperation. The desirable effect of exercise on plasma cholesterol profile is strengthened in adult age than young age.

Key words: vitamin B_6 defidiency, age, cholesterol profile, exercise

INTRODUCTION

When the body is involved in physical exertion, certain metabolic processes occur to assure adequate energy is provided to the exercising muscles of the body and is associated with variable effects on the blood cholesterol profile. The effect of vitamin B_6 deficiency on cholesterol metabolism during exercise remains controversial. Leklem(1) reported that vitamin B_6 deficiency was not associated with the significant change in serum cholesterol. Song and Cho(2) reported that the level of plasma total cholesterol was positively correlated with the level of plasma vitamin B_6 in healthy men. However it is also reported that the plasma pyridoxal 5'phosphate(PLP) levels are positively correlated with plasma high density lipoprotein(HDL)-cholesterol levels and negatively correlated with total cholesterol and low density lipoprotein(LDL)-cholesterol levels in monkeys (3). Studies in vitamin B_6 deficient rats showed there was an accumulation of lipid, consisting mainly of triglyceride and cholesterol(4-7).

Aerobic exercise increased HDL-cholesterol in moderately exercised obese women(8) as well as in heavily exercised men(9), while aerobic exercise had little or no effect on triglyceride and LDL-cholesterol unless combined with body weight losses in obese men and women(8,10,11). At present, there are no definitive studies on the effect of vitamin B_6 deficiency on plasma cholesterol level during exercise. It is generally known that age affects the cholesterol metabolism. Also, vitamin B_6 dificiency is common in the elderly due to poor dietary intake. Neverthless, there is strong evidence that aging affects the reqirement of vitamin B_6 and metabolite assays permit identification of elderly subjects who may benefit from vitamin supplementation(12-14).

Therefore, the aims of this study were as follows: 1) to determine whether cholesterol metabolism was affected by vitamin B_6 deficiency during exercise and 2) to determine whether plasma cholesterol profile was affected by age during exercise.

MATERIALS AND METHODS

Diets and exercise

A total of 54 weanling male Sprague-Dawley rats were received either vitamin B_6 deficient($-B_6$) diet or control($+B_6$) diet. Animals received a vitamin-free, casein-based semisynthetic diet which met AIN-76 recommendations(15,16) with the exception of vitamin B_6. The diet contained by weight 20% protein, 5% fat and 65% carbohydrates. These rats were fed for 6 weeks. At week 3, eighteen rats from control group were sub-divided into 3 groups; non-exercise group(NE), exercise and sacrifice group(ES), exercise and recuperation group(ER). ES group was exercised on treadmill(10, 0.5~0.8km/h) for 2 hours and sacrificed right after exercise. ER group was recuperated for three days with respective diet. At week 6, 36 rats with respective diet were subdivided, exercised same as that of 3 weeks.

At the respective time points(non-exercise, right after 2 hours exercise, 3 days recuperation after exercise), animals were sacrificed by decapitation under light ether anesthesia after 16 hours fasting. Immediately following decapitation, blood was collected in heparinized tubes and centrifuged to separate plasma. Plasma was stored at -40°C until analyzed.

Biochemical and statistical analysis

Plasma total cholesterol(TC) was analyzed with commercial kit utilizing cholesterol oxidase(Youngdong Pharmaceutical Co. Korea), HDL-cholesterol was analyzed with commercial kit based on same analytical method as total cholesterol, after the precipitation of LDL, very low density lipoprotein(VLDL) and chylomicron with polyethyleneglycol(International Reagent Co., Japan). Triglyceride(TG) was analyzed with commercial kit based on Trinder method(Youngdong Pharmaceutical Co., Korea). LDL-cholesterol was estimated by the method of Friedwald et al.(17).

All data were evaluated by analysis of variance. For those F values which were significant, Duncan's multiple range test(18) was performed. A p value<0.05 was considered to be significant.

RESULTS

The effects of vitamin B_6 deficiency on plasma total cholesterol level in exercising rats is shown in Fig. 1. In NE group, there was no difference in plasma total cholesterol between $+B_6$ and $-B_6$ rats($+B_6$, 110.9±13.47; $-B_6$, 109.68±30.38, mg/dl). Although the level of plasma total cholesterol in ES group($+B_6$, 93.7±6.65; $-B_6$, 96.75±18.63, mg/dl) or ER group($+B_6$, 98.85±16.75 ; $-B_6$, 85.85±25.09, mg/dl) tended to be lower than that of NE group, there was also no difference between $+B_6$ and $-B_6$ rats in ES group or ER group.

Fig. 2 shows the effects of vitamin B_6 deficiency on plasma HDL-cholesterol level in exercising rats. In NE group, there was no difference between $+B_6$ rats and $-B_6$ rats in plasma level of HDL-cholesterol($+B_6$, 36.85±6.35; $-B_6$, 36.68±7.25, mg/dl). Compared to NE group, this level tended to be lower in ES group of both $+B_6$ and $-B_6$ rats although the differences were not statistically significant due to the large standard deviation($+B_6$, 31.43±5.68; $-B_6$, 28.48±6.78, mg/dl). In ER group, the level of plasma HDL-cholesterol increased and returned to that of NE group in $+B_6$ rats, while the level of $-B_6$ rats was not increased and was significantly lower than that of $+B_6$ rats($+B_6$, 38.48±8.47; $-B_6$, 24.94±6.72, mg/dl).

Fig. 3 shows the effect of vitamin B_6 deficiency on plasma LDL-cholesterol level in exercising rats. The tendency of plasma LDL-cholesterol change due to

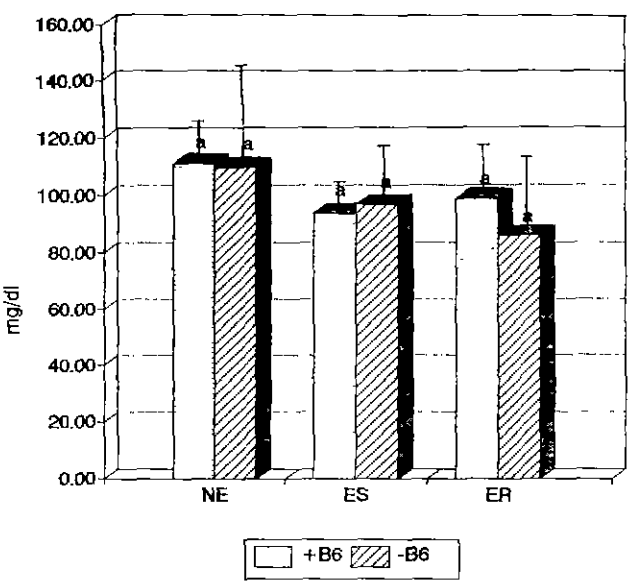

Fig. 1. The effect of vitamin B_6 deficiency on plasma total cholesterol level during exercise.
Each bar with different letters is significantly different($p<0.05$): $+B_6$=control diet pair fed to $-B_6$ group; $-B_6$=vitamin B_6 deficient diet; NE=non-exercise; ES=sacrifice right after two hours exercise; ER=two hours exercise and recuperation for three days

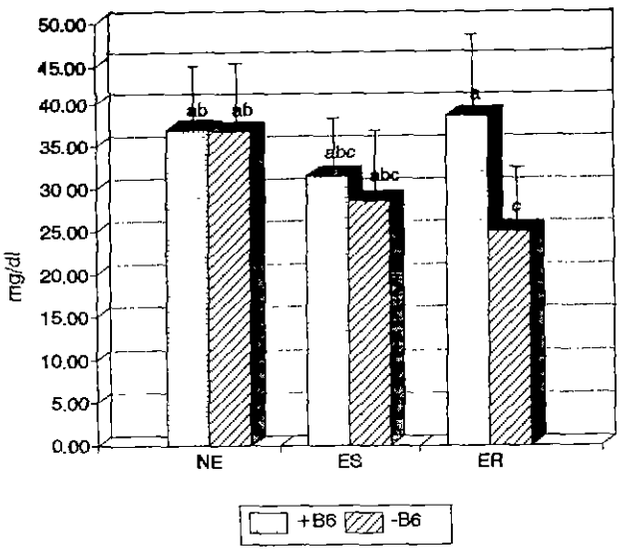

Fig. 2. The effect of vitamin B_6 deficiency on plasma high density lipoprotein-cholesterol level during exercise.

Each bar with different letters is significantly different($p<0.05$): $+B_6$=control diet pair fed to $-B_6$ group; $-B_6$=vitamin B_6 deficient diet; NE=non-exercise; ES=sacrifice right after two hours exercise; ER= two hours exercise and recuperation for three days

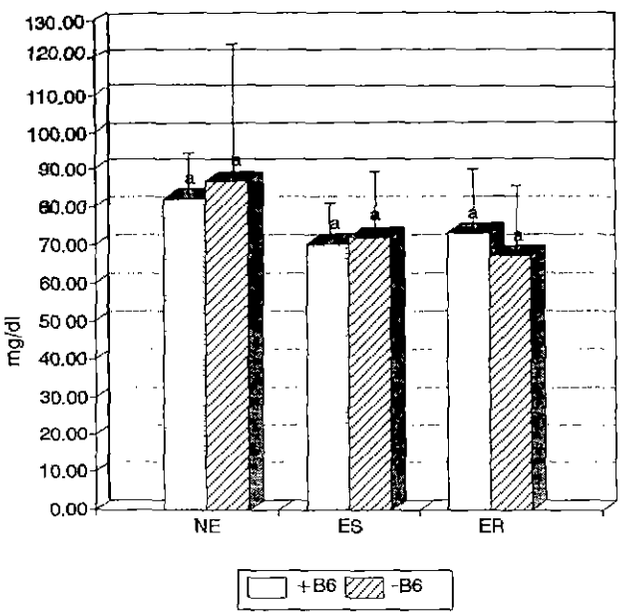

Fig. 3. The effect of vitamin B_6 deficiency on plasma low density lipoprotein-cholesterol level during exercise.

Each bar with different letters is significantly different($p<0.05$): $+B_6$=control diet pair fed to $-B_6$ group; $-B_6$=vitamin B_6 deficient diet; NE=non-exercise; ES=sacrifice right after two hours exercise; ER= two hours exercise and recuperation for three days

vitamin B_6 deficiency and exercise or recuperation was similar to that of total cholesterol, ie. no difference in LDL-cholesterol was shown between $+B_6$ and $-B_6$ rats or among NE($+B_6$, 82.05±9.35; $-B_6$, 86.65±35.03, mg/dl), ES($+B_6$, 70.15±7.63; $-B_6$, 71.83±14.49, mg/dl)or ER($+B_6$, 73.00±13.67; $-B_6$. 67.24±15.46, mg/dl) group.

The effect of age on plasma total cholesterol level in exercising rats is shown in Fig. 4. In NE group, the total cholesterol level of 6 weeks was significantly higher than that of 3 weeks rats(3wk, 86.7±22.7; 6wk, 110.9±13.4, mg/dl). However, this level of 6 weeks rats was decreased in ES group(3wk, 93.7±19.7; 6wk, 93.7±6.7, mg/dl) and there was no difference between 3 weeks and 6 weeks rats in ES group. In ER group, the level of plasma total cholesterol was increased significantly in 3 weeks rats but this level was not increased in 6 week rats(3wk, 110.6±24.9; 6wk, 98.9±16.8, mg/dl).

Fig. 5 shows the effects of age on plasma HDL-cholesterol level in exercising rats. In NE group, there was no difference in the level of HDL-cholesterol between 3 weeks and 6 weeks(3wk, 38.11±7.7; 6wk, 36.9±6.4, mg/dl). In ES group, the HDL-cholesterol level of 3 weeks rats was not changed, while that level of 6 weeks rats was decreased and significantly lower than that of 3 weeks rats(3wk, 40.8±9.3; 6wk, 31.4±5.7, mg/dl). In ER group, the plasma HDL-cholesterol level of 3 weeks rats was lower than that of 6 weeks rats (3wk, 29.3±3.1; 6wk, 38.4±8.4, mg/dl).

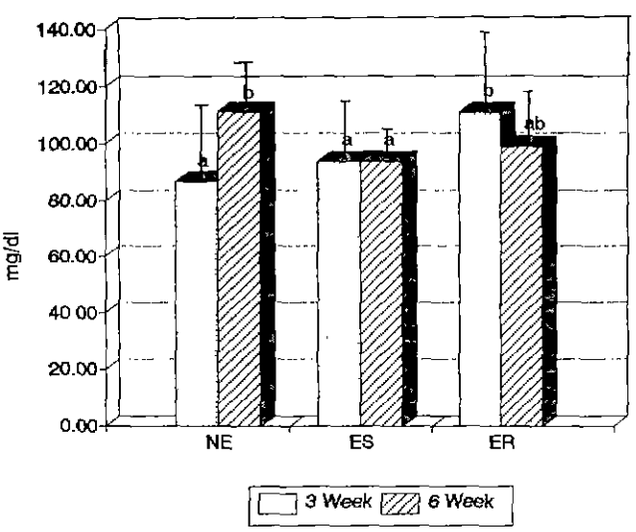

Fig. 4. The effect of age on plasma total cholesterol level during exercise.

Each bar with different letters is significantly different($p<0.05$): 3 week=3 weeks of age after weanling; 6 week=6 weeks of age after weanling; NE=non-exercise; ES=sacrifice right after two hours exercise; ER= two hours exercise and recuperation for three days

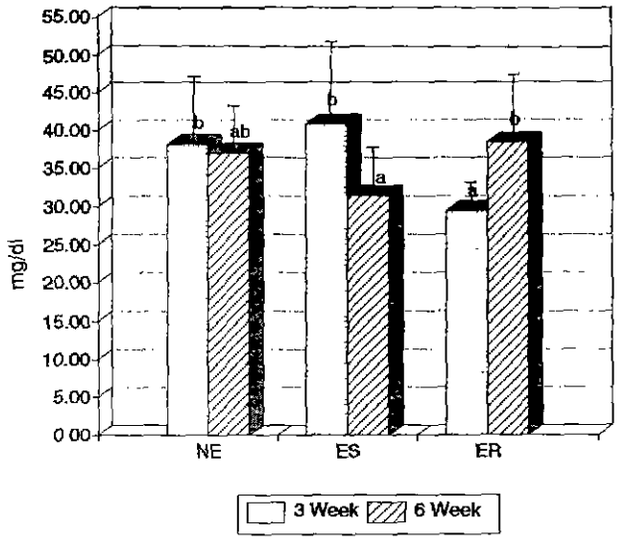

Fig. 5. The effect of age on plasma high density lipoprotein-cholesterol level during exercise.
Each bar with different letters is significantly different($p<0.05$): 3 week=3 weeks of age after weanling; 6 week=6 weeks of age after weanling; NE=non-exercise; ES=sacrifice right after two hours exercise; ER= two hours exercise and recuperation for three days

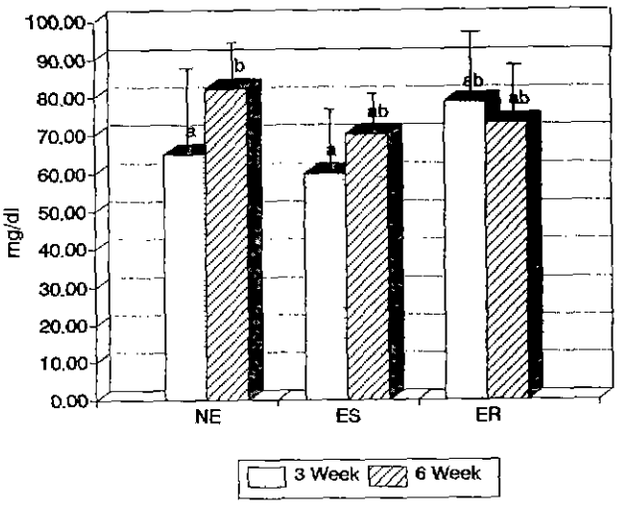

Fig. 6. The effect of age on plasma low density lipoprotein-cholesterol level during exercise.
Each bar with different letters is significantly different($p<0.05$): 3 week=3 weeks of age after weanling; 6 week=6 weeks of age after weanling; NE=non-exercise; ES=sacrifice right after two hours exercise; ER= two hours exercise and recuperation for three days

Fig. 6 shows the effects of age on plasma LDL-cholesterol level in exercising rats. In NE group, the LDL-cholesterol level of 6 weeks rats was significantly higher than that of 3 weeks rats(3wk, 64.9±20.2; 6wk, 82.1±9.4, mg/dl). In ES group, this level of 6 weeks tended to be higher than that of 3 weeks rats although the difference was not signficant(3wk, 60.0±14.1; 6wk, 70.2±7.6, mg/dl). In ER group, there was no differences of plasma LDL-cholesterol level between 3 weeks rats and 6 weeks rats(3wk, 78.5±15.99; 6wk, 73±13.7, mg/dl).

DISCUSSION

The findings that plasma total cholesterol levels remained unchanged in $-B_6$ rats were consistent with the previous reports(19-21). But these results were not consistent with those of Suzuki and Okada(7); the fall in total cholesterol followed a fall in phospholipid in plasma of vitamin B_6 animals. A possible reason for this discrepancy may be due to the type of diet(70% casein vs 20% casein). It has been reported that only regular moderates exercise training increases HDL-cholesterol and decreases LDL-cholesterol and triglyceride(22-24). However, in this study, even an abrupt intense exercise tended to decrease HDL-cholesterol in both $+B_6$ rats and $-B_6$ rats although the difference was not significant. Because it has been reported that plasma PLP is elevated during endurance exercise and leads net loss of vitamin B_6 result from the urinary increase of 4-pyridoxic acid (25,26), exercise can aggravate vitamin B_6 deficiency. Thus, the deteriorative effect on the plasma HDL-cholesterol level due to the insufficient amount of PLP might be showed in $+B_6$ rats and $-B_6$ rats during exercise and $-B_6$ rats after recuperation. The another support for this is from the result that HDL-cholesterol level of $+B_6$ rats returned to the level of non exercised group after recuperation. Nonetheless, the mechanism which decrease the HDL-cholesterol level during exercise could not be entirely identified from the results of this study.

It has been reported that the levels of blood total cholesterol and LDL-cholesterol were elevated with age(27,28). The result of this study is consistent with those reports in non-exercised group. However, during exercise, 3 weeks rats did not change the blood level of total cholesterol and LDL-cholesterol, while 6 weeks rats decreased those levels and they became to show the same levels as that of 3 weeks. Even after recuperation, the plasma levels of total cholesterol and LDL-cholesterol of 6 weeks rats was not higher than those of 3 weeks rats. Thus, even an abrupt exercise has

more desirable effect on blood level of total cholesterol and LDL-cholesterol in 6 weeks than in 3 weeks. The implication of lower HDL-cholesterol in 6 weeks compared to 3 weeks during exercise found in this study is unclear. The mechanism involved here might indicates that there is no need to excrete cholesterol from the tissues because of the lowered total cholesterol and LDL-cholesterol during exercise in 6 weeks rats, or these animals were unable to secrete lipoproteins from liver into circulation(29,30).

Thus, it is concluded that vitamin B_6 deficiency may affect the blood cholesterol profile especially on HDL-cholesterol during exercise and after recuperation. The desirable effect of exercise on plasma cholesterol profile is strengthened in adult age rather than young age.

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