# High-Density Lipoprotein Cholesterol Changes after Continuous Egg Consumption in Healthy Adults

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**Objective:** To determine the relationship between continuous egg consumption with Thai life-style dietary and serum lipids of healthy young people.

*Material and Method:* Fifty-six participants with an average age of 35 were enrolled. In an experimental method of cholesterol intake, all participants were fed an additional egg per day to their basic diet. This project ran for 12 weeks.

**Results:** The 12-week egg consumption significantly increased serum total cholesterol by  $0.27 \pm 0.15$  mmol/L  $(10.43 \pm 5.80 \text{ mg/dL})$  (p < 0.05). The HDL-cholesterol (HDL-c) increased significant by  $0.55 \pm 0.06$  mmol/L  $(21.80 \pm 2.25 \text{ mg/dL})$ (p < 0.001) while the total cholesterol (TC) decreased as the HDL-c ratio was  $0.94 \pm 1.1$  (p < 0.001). No significant changes were found in LDL-cholesterol (LDL-c) and triglyceride levels. The present study showed that small serum LDL-c changed in response to change of egg consumption. Additionally, 12-week egg consumption also resulted in an increasing HDL-c level.

**Conclusion:** In the majority of healthy adults, an addition of one egg per day to a normal fat diet could raise HDL-c levels and decreased the ratio of TC to HDL-c. Therefore, egg consumption might benefit blood cholesterol level.

Keywords: Cholesterol, Egg consumption

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High serum levels of low-density lipoprotein cholesterol (LDL-c) are a major risk factor of coronary heart disease (CHD)<sup>(1)</sup>. To avoid elevating blood cholesterol and reducing CHD risk, the public have been advised to consume no more than 300 mg/d of cholesterol and to limit consumption of eggs no more than 3 to 4 whole eggs a week since 1970. Over the years there have been numerous reports that egg consumption was not related to either serum cholesterol levels or coronary heart disease incidence. This is because it raised level of total cholesterol but not LDL-c in blood compared with saturated and *Tran* fatty acid<sup>(1-8)</sup>. Several studies revealed that dietary cholesterol

increased not only concentrations of LDL-c but also concentrations of HDL-cholesterol (HDL-c)(26). However, other studies found that individuals vary widely in their responses to dietary cholesterol by many factors as cholesterol in the diet was not the principal factor affecting the level of cholesterol in the bloodstream<sup>(9)</sup>. Besides, different research designs also illustrated different outcomes such as researches conducted in free-living population<sup>(9,10)</sup> and those in control studies<sup>(6-8)</sup>. A new scientific study completed in 1996 confirmed and strengthened the conclusions that dietary cholesterol had only a small effect on blood cholesterol<sup>(12)</sup>. But the authors know that blood cholesterol is affected by many mechanisms including race, dietary life-style and culture. In 1999, Hu et al<sup>(2)</sup> reported a correlation of a large prospective cohort study of egg consumption and risk of cardiovascular

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disease in men and women, and the finding showed that there was no evidence of a significant relationship between egg consumption and CHD or stroke in both men and women.

Restricting eggs from the diet could have negative nutritional effects. Eggs are the largest single source of cholesterol among the foods commonly eaten in the world; a large egg contains about 213 mg of cholesterol<sup>(11)</sup>. However, eggs also contain many other nutrients besides cholesterol<sup>(28)</sup>, including unsaturated fats, essential amino acids, iron, folate, vitamins-B, -D, -E and the best sources of an important nutritional substance called Choline. If people eliminate or reduce specific food (e.g., eggs) from their diet, they may replace it with calories from another source. It is altogether possible that the food that replaces eggs is higher in calories and/or fat.

In Thailand, the authors have not found a published study about egg consumption and blood cholesterol since dietitians still recommended the public to restrict egg consumptions in no more than four whole eggs a week. In the present research, the authors prospectively studied the association between egg consumption with a Thai life-style controlled diet and blood cholesterol levels in 56 Thai volunteers.

# **Material and Method**

Fifty-six participants (51 men and 5 women) were recruited from a population of 80 workers from the Industrial Rehabilitation Center (IRC), Ministry of Labor. The participants were between 20 and 50 years old, and agreed to work and stay in the IRC throughout the study. They had their blood test levels tested by T.A. Central Lab Co., Ltd. Blood chemistry; consisting of Total cholesterol (TC), LDL-c, HDL-c, and Triacyl-glycerol (TG). Questionnaires were used to screen the participants.

The coordinating dietitians interviewed all the participants before they were enrolled regarding daily dietary intake, ethanol consumption, and smoking habits. Individuals with extreme dietary habits, overweight (BMI > 25) or with significant food intolerances were excluded. Moreover, serum TC levels were less than 240 mg/dL. The authors also excluded participants who smoked or followed vigorous exercise regimens. None had serious medical problems, recent laboratory testing abnormalities (such as hyperglycemia, hypertension (> 140/90 mmHg), CHD, or cerebrovascular disease) or were continuously taking medications that might affect serum lipid levels were excluded. All the participants were in good health. Having had a similar life-style, all the participants had stayed at IRC in training programs for at least four months. Furthermore, most of them had eaten the same three meals.

The experimental protocol was reviewed and approved by the committee of Institute of Health Research, Chulalongkorn University. They were informed in detail about this research and informed consent was obtained from all participants.

# Protocol

This study was designed to determine the response of young adults who added one whole egg per day to their basic diet as recommended by the American Heart Association (AHA) and the National Cholesterol Education Program (NCEP).

Six blood samples, of 6 milliliters each, were taken twice a month for determination of lipids. All fasting samples were obtained between 8 and 9 AM after a 12-hour overnight fast.

#### Diets

All meals were prepared with fresh ingredients. The participants were served with a variety of food such as beef, pork, fish, fruits, vegetables, noodles, legumes, and desserts. Every subject ate the same kind of diet as he/she usually did, but would be individually different in amount of the diets. A 4-week menu cycle was used throughout the present study. The composition of the research menus was the same as in previous months and based on NCEP<sup>(3)</sup>. It contained 50-60% of calories from carbohydrates, 15% from protein, and 25-35% from total fat, which contained less than 7% in saturated fat. The egg was added by weight (approximate weight, 55 grams), which, according to Thailand's recommended dietary intakes (Thai RDIs)<sup>(4)</sup>, contains approximately 213 mg cholesterol each. The egg was served daily at lunch. Each day, egg menus were different with a 6-egg-menu cycle (egg boiled, omelet, fried egg, soft-boiled egg, egg stewed in the gravy, and stuffed omelet).

The 24-hour dietary records done by all participants used to determine the average daily intake of energy and certain nutrients (carbohydrate, total protein, fat, dietary cholesterol and calories). Those records were done every week of the 12-week study, 3-days a week (Sunday, Monday and Tuesday) and were recalled again by coordinating dietitians, Department of Nutrition, Mahidol University on every Tuesday. The authors used a 3-day dietary record<sup>(29,30)</sup> to represent the average total energy intake and certain

nutrients of those weeks.

#### Laboratory

Trained assistants used standardized techniques to measure blood pressures during the present study. Height and weight were measured every month by using manual techniques. TC, TG HDL-c and Glucose concentrations were measured by enzymatic techniques with an automatic serum analyzer, Hitachi 717 (Boehringer Mannheim, GmbH, Mannheim, Germany) to obtain fasting, intravenous blood chemistry levels. Using reagents supplied by S.E. Supply Co. and E for L Co, the serum LDL-c was calculated according to the Friedewald formula<sup>(5)</sup>.

# Statistical analysis

Demographic and baseline characteristics (including age, sex, education, occupation, and income) of all participants were assessed by using mean + standard error mean (SEM). Average total energy intake and certain nutrients were also assessed and analyzed using an analysis of variance (ANOVA). For all serum lipid profiles (TC, TG, HDL-c, LDL-c), fasting blood glucose - the authors used mean concentrations of TC, and HDL-c at the end of each diet to estimate the mean ratios of TC to HDL-c concentrations, an analysis of variance was used to determine whether there was a significant effect of egg consumption on the parameters determined. Statistical analysis was performed on the data between baseline (t=0) and each period (a paired two-tail t-test was used to compare data obtained at the beginning and the end of each blood collection). All statistical analyses of the lipid responses were done with adjusted values by using SPSS for Windows version 11.0. Point estimates and 95% confidence intervals were calculated for the mean differences in change between each.

## Results

Table 1 presents the baseline characteristics of the 56 participants with the mean age of  $35.64 \pm 1.47$ years, 91.1% of male, and 8.9% of female. Body weight remained constant throughout the present study. Some participants did not completely record 24-hour dietary records in some weeks. At the end of the present study, ten of the original 56 participants withdrew because their training was completed at the Rehabilitation center, and 2 participants did not complete blood collecting in the 6<sup>th</sup> week because they went to visit their family. The mean  $\pm$  SEM for all participants baseline blood sugar and lipids are presented in Table 3. The analyzed and calculated compositions of nutrients are presented in Table 2. The authors had found that the compositions of the diets between  $1^{st}$ ,  $2^{nd}$ , and  $3^{rd}$  month were not changed except significant changes of dietary protein between  $1^{st}$  and  $2^{nd}$  month. Cholesterol intakes had no changes for the entire study. The quantities of total fat, carbohydrate, and protein approximately met the guidelines of the NCEP diet.

# Effects to blood lipid levels

The authors recorded the serum lipid levels at the beginning of the present study,  $2^{nd}$  week,  $4^{th}$ week,  $6^{th}$  week,  $10^{th}$  week and  $12^{th}$  week. Results of egg consumptions and blood lipid levels are shown in Table 3 and Fig. 1. The authors found that serum total Cholesterol significantly increased (p < 0.05) equal to 4.71 mmol/L. Moreover, HDL-c and TC: HDL-c ratio both significantly changed (p < 0.001). HDL-c increased by 48.38 percent, while TC: HDL-c ratio decreased by 25.82 percent with no statistically significant changes in TG and LDL-c levels.

#### Discussion

The authors sorted previous experimental studies into two groups. Those with metabolic ward studies, in which all food was provided<sup>(6-8, 31)</sup>, and those with free-living subjects that were provided eggs, high cholesterol products, or egg-free substitutes<sup>(9-10,32)</sup>. Both methods showed advantages and disadvantages. In the free-living method, the findings may vary simply because subjects could consume different types of food, while the food-control study could solve this complication. However, although every subject ate the same type of food provided daily by the researcher, the type of food itself changed everyday causing changes in dietary cholesterol level.

**Table 1.** Summary of demographics and baseline characteristics of all participants (mean  $\pm$  SEM)

Characteristics	
Sex: male/female	51/5
Age (year)	$35.60 \pm 1.47$
Drink/no drink	60.7/39.3
Smoke/no smoke	53.6/46.4
Weight (kg)	57.20 <u>+</u> 1.13
Body mass index (kg/m <sup>2</sup> )	20.90 <u>+</u> 0.39
Systolic blood pressure	$111.45 \pm 1.41$
Diastolic blood pressure	$71.18 \pm 1.03$

SEM; standard error mean

Table 2.	Mean + SEM	of the average d	lietary intakes (	(per day)

	First month Mean $\pm$ SEM	$2^{nd}$ month Mean $\pm$ SEM	$3^{rd}$ month Mean $\pm$ SEM
Total energy intake (kcal) Carbohydrate (g)	$\begin{array}{c} 1,780.9 \pm 40.72 \\ 269.7 \pm 6.76 \end{array}$	$\begin{array}{c} 1,777.2 \pm 34.76 \\ 272.7 \pm 5.91 \end{array}$	$\begin{array}{c} 1,761.4 \pm 25.09 \\ 265.1 \pm 4.75 \end{array}$
Protein (g)	53.3 <u>+</u> 1.35	$\pm 1.35$ 51.4 $\pm 0.87^*$ 52.4	$52.4 \pm 0.81$
Fat (g)	53.8 <u>+</u> 1.43	54.0 <u>+</u> 1.53	$55.6 \pm 0.98$
Cholesterol (mg)	354.6 <u>+</u> 9.05	351.4 <u>+</u> 5.83	367.1 <u>+</u> 8.66

\* Significant difference from first month at p < 0.05

All means were compared with means in first month. All means were the average of each month

**Table 3.** Mean  $\pm$  SEM changes in serum lipid concentrations at each level of dietary cholesterol

	Control	2 <sup>nd</sup> wk	$4^{th} wk$	6 <sup>th</sup> wk	$8^{th} wk$	12 <sup>th</sup> wk
TC (mg/dL) TG (mg/dL) HDL-c (mg/dL) LDL-c (mg/dL) TC:HDL-c	$\begin{array}{c} 4.36 \pm 0.98 \\ 1.23 \pm 0.47 \\ 1.20 \pm 0.41 \\ 2.63 \pm 0.82 \\ 3.64 \pm 2.41 \end{array}$	$\begin{array}{c} 5.22 \pm 0.88^{**} \\ 1.38 \pm 0.92 \\ 1.10 \pm 0.16^{**} \\ 3.47 \pm 0.7^{**} \\ 4.85 \pm 1.02^{**} \end{array}$	$\begin{array}{c} 5.02 \pm 1.35^{**} \\ 0.89 \pm 0.46^{**} \\ 1.91 \pm 0.32^{**} \\ 2.90 \pm 0.86^{*} \\ 2.69 \pm 0.81^{**} \end{array}$	$\begin{array}{c} 5.08 \pm 0.98^{**} \\ 1.04 \pm 0.72^{*} \\ 2.03 \pm 0.25^{**} \\ 2.58 \pm 0.84 \\ 2.53 \pm 0.54^{**} \end{array}$	$\begin{array}{c} 4.85 \pm 1.02 * \\ 1.37 \pm 0.63 \\ 1.70 \pm 0.19 * * \\ 2.50 \pm 0.99 \\ 2.88 \pm 0.68 * * \end{array}$	$\begin{array}{c} 4.75 \pm 1.07 * \\ 1.18 \pm 0.59 \\ 1.78 \pm 0.19 * * \\ 2.44 \pm 0.98 \\ 2.70 \pm 0.65 * * \end{array}$

\* Significant difference from First month at p < 0.05

\*\* Significant difference from First month at p < 0.001

TC, total cholesterol; HDL-c, High-density lipoprotein cholesterol; LDL-c, low-density lipoprotein cholesterol;

TC:HDL, ratio of total cholesterol to HDL-cholesterol

To convert mmol/L of HDL or LDL cholesterol to mg/dL, multiply by 39

To convert mmol/L of triglycerides to mg/dL, multiply by 89

All means were compared with means in first month. All means were the average of each month



Fig. 1 Shows serum lipid at each time

Ginsberg et al<sup>(11)</sup> investigated the effect of increasing dietary cholesterol in 13 female medical and dental students. They all ate under supervision of the coordinating dietitian. All menus were prepared. The food may not have been the same type of food that subjects normally consumed in everyday life. However,

in the present study, the authors used before-and-after design and food menus that the subjects had regularly consumed in the previous four months to observe changes that could be fully attributed to the change in egg consumption. This eliminated the drift of variables over time. Together with having a significant number of subjects - 56 participants, this study should be able to support the relationship between egg consumption and dietary cholesterol blood level. There were no statistical changes in total energy intakes and nutrients along the project. Many researchers have published the conclusion about Acceptable Macronutrient Distribution Ranges (AMDR, 2002) that adults should consume nutrients between 20-35% of fat composition, 45-65% of carbohydrate composition, and 10-35% of protein composition. To prevent the exceeding consumption of fat composition, people should not consume carbohydrate less than 45%. In the present study, the authors found that the participants consumed a favorable range of nutrients. The participants consumed high carbohydrate (about 60%) that came from rice and flour, 15% of proteins, and 22-28% of fat. The total energy intakes were about 1,800 to 2,000 kcal. As for cholesterol, the important nutrient, the authors were particularly interested because the participants consumed these nutrients, 354-400 mg per day, which was more than the recommendation of NCEP. The suggestion from NCEP is to limit consumption of cholesterol at 300 mg per day.

The results of the present study indicated that the addition of one egg per day to a normal fat diet had little effect on serum LDL-c levels, similar to many studies. However, there were important differences between the present study and other previous published studies. The serum TC and HDL-c significantly increased by 6.67% and 54.81%, respectively. Moreover, ratios of TC to HDL-c significantly decreased (p < 0.001), but had no statistical changes in LDL-c and TG levels. Similar to the study of Schnohr (1994), after six weeks of extra egg consumption, the serum HDL-c increased by 10% (p < 0.05), the TC increased by 4% (p < 0.05), and the ratio TC to HDL-c did not change significantly. The reason of the increase in TC was simply because of the high increase in serum HDL-c levels that the body had responded after consuming a higher cho-lesterol nutrient. The increase of HDL-c and decrease of ratio TC to HDL-c in these participants were good results similar to other studies such as the Framingham Heart Study<sup>(3,12)</sup>, the Multiple Risk Factor Intervention Trial (MRFIT)<sup>(13)</sup>, the Lipid Research Clinics Prevalence Trial<sup>(14)</sup>, the Alpha-Tocopheral, Beta-Carotene Cancer Prevention Study(15), the Nurses' Health Study<sup>(16)</sup>, and the Health Professionals Follow-Up Study<sup>(17)</sup>.

Most studies on the effects of dietary cholesterol on serum lipids were less than 10 weeks and the number of subjects per study ranged from 9 to 131. Ginsberg et al<sup>(6)</sup> investigated the effects of dietary cholesterol in 24 healthy young men, which used four diet periods of 8 weeks each to examine the effects of addition of zero (128 mg cholesterol/day), one (283 mg/ day), two (468 mg/day), or four (858 mg/day) eggs per day to the basic diet. However, these study results were similar to the authors' findings that the addition of two eggs per day to a low-fat diet had little effect on serum cholesterol levels. However, the present study had a significant increase in HDL-c, similar to the findings Clifton and Nestel<sup>(18)</sup> demonstrated. They had a greater increase in HDL-c in women versus men. Those findings were consistent in both age groups analyzed.

Many epidemiological surveys have often found a negative relationship between dietary cholesterol and CHD mortality when saturated fat and dietary fiber were included in the analysis. Moreover, in 1997, Hu et al<sup>(16)</sup> published an article indicating types of fat that had a more important role in determining risk of CHD than the total amount of fat in diet. Many studies have shown that saturated fats and trans-fats have a greater impact than did dietary cholesterol in raising blood cholesterol levels. Diets high in saturated fatty acids raised serum LDL-c levels. Reduction in intakes of saturated fatty acids lowered LDL-c levels and reduces risk for CHD<sup>(22-27)</sup>.

As to the previous knowledge, there were good fats and bad fats. In the public's mind, fat had become the number one public enemy. Reducing dietary fat had become a priority, but the truth lay in the fact that if one reduces his/her total fat consumption, one will also reduce the amount of good fats that have a protective effect against heart disease. Therefore, rather than worry about the number of dietary eggs consumed per day, one would rely on the type of dietary fats.

The exact reason of decreasing serum TC and LDL-c has not been authenticated. However, the authors believe that the substance compound in egg volk and lecithin had a hypo-cholesterolemic effect -Lecithin belongs to a group of nutrients known as lipids (fats, oils, waxes) and was a phospholipid called phosphatidyl choline. Because of Lecithin, the authors did not observe hypocholesterolemic effects from the previous researches comparing blood cholesterol with dietary of other food containing cholesterol. The second reason was that blood cholesterol response was independent to baseline blood cholesterol level and dietary food types of Thai life-style, which usually had more vegetables, legumes, and other essential nutrients. In the present study, the hypocholesterolemic effect may come from the combination of other Thai life-style dishes and lecithin-containing eggs.

In conclusion, the present study demonstrated that, for the majority of healthy young people an addition of one egg per day to a normal fat diet could raise the level of HDL-c and decrease the ratio of TC to HDL-c. Egg consumption may benefit blood cholesterol level.

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# ระดับ HDL-cholesterol เพิ่มขึ้นในผู้ใหญ่สุขภาพดีที่รับประทานไข่

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**บทนำ**: เป็นที่ทราบกันดีว่า การเพิ่มของระดับคอเลสเตอรอลในเลือด เป็นปัจจัยเสี่ยงของโรคหลอดเลือด ทำให<sup>้</sup>ตลอด 30 ปีที่ผ่านมาเกิดคำแนะนำให้คนทั่วไป ลดการรับประทานอาหารที่เป็นจำพวกไขมัน รวมทั้งไข่ด้วย และคำแนะนำนี้ ส่งผลให้ประชาชนโดยเฉพาะเด็กรุ่นใหม่หลีกเลี่ยงการรับประทานไข่ ทั้ง ๆ ที่เป็นแหล่งโปรตีนและสารอาหารต่าง ๆ ที่มีคุณค่าต่อร่างกาย

**วัตถุประสงค**์: เพื่อศึกษาระดับไขมันในเลือด และวัดการเปลี่ยนแปลงของระดับไขมันในเลือดหลังจากการรับประทานไข่ อย่างต่อเนื่องจำนวน 1 ฟองต่อวันเป็นเวลา 12 สัปดาห์ ในคนไทยวัยทำงานสุขภาพดี

**วัสดุและวิธีการ**: ศึกษากับประชากร ที่ศูนย์พื้นฟูอาชีพ สำนักงานประกันสังคม จำนวนทั้งหมด 56 คนตามลำดับ อายุเฉลี่ยประมาณ 35 ปี โดยเป็นการศึกษาเชิงทดลองที่มีการบันทึกสารอาหารที่ได้รับต่อวัน โดยใช้แบบบันทึกอาหาร ที่บริโภค เป็นเวลา 3 วันต่อสัปดาห์ (24-hour dietary record) และมีการตรวจเลือดอย่างต่อเนื่องจำนวน 6 ครั้ง

**ผลการศึกษา**: พบว่าหลังจากรับประทานไข่ติดต่อกัน 12 สัปดาห์ ระดับ Total Cholesterol(TC) มีค่าที่เพิ่มขึ้นอย่าง มีนัยสำคัญทางสถิติ โดยเพิ่มขึ้น 0.27 <u>+</u> 0.15 mmol/L (10.43 <u>+</u> 5.80 mg/dL) (p < 0.05) ซึ่งเนื่องจาก HDL-cholesterol (HDL-c) ที่มีค่าเพิ่มขึ้นอย่างมีนัยสำคัญทางสถิติ โดยเพิ่มขึ้น 0.55 <u>+</u> 0.06 mmol/L (21.80 <u>+</u> 2.25 mg/dL (p < 0.001) สำหรับอัตราส่วนระหว่าง TC ต่อ HDL-c ลดลงอย่างมีนัยสำคัญทางสถิติ โดยลดลง 0.94 <u>+</u> 1.11, p < 0.001) ส่วน LDL-Cholesterol (LDL-c) และ Triglyceride นั้นไม่มีความแตกต่างกัน (p > 0.05) ผลการทดลองพบว่าการบริโภคไข่ ไม่ได้ทำให้ระดับ LDL-c สูงขึ้น ยิ่งไปกว่านั้นการรับประทานไข่ในระยะยาว 12 สัปดาห์กลับทำให้ระดับ ไขมัน HDL-c สูงขึ้นมาก

**สรุป**: การรับประทานไข่ ติดต่อกัน 12 สัปดาห์ในคนวัยทำงานสุขภาพดีกลุ่มนี้ ไม่ทำให้ระดับระดับโคเลสเตอรอลในเลือด LDL-c สูงขึ้นแต่กลับทำให้ระดับโคเลสเตอรอลHDL-c สูงขึ้นได้ด้วย