Indian J Physiol Pharmacol 2004; 48 (3): 286–292

THE EFFECT OF INGESTION OF EGG ON THE SERUM LIPID PROFILE OF HEALTHY YOUNG INDIANS

GAYATRI CHAKRABARTY, S. MANJUNATHA, R. L. BIJLANI*, ROOMA BASU RAY, S. C. MAHAPATRA, NALIN MEHTA, R. LAKSHMY**, SUMAN VASHISHT** AND S. C. MANCHANDA**

*Departments of Physiology & **Cardiology, All India Institute of Medical Sciences, New Delhi – 110 029*

(Received on March 16, 2004)

Abstract : Thirty four healthy young volunteers (22 men, 12 women; age 25.7±5.8 years; BMI 20.8±2.3 kg/m^2) participated in a randomized controlled cross-over trial on the effect of consuming one boiled egg every day for 8 wk on the serum lipid profile. The only significant change after 8 wk of egg consumption was an elevation of the total cholesterol/HDL cholesterol ratio. However, scrutiny of individual responses revealed that twelve of the subjects (10 men, 2 women) had a greater than 15% rise in the LDL cholesterol level after 8 wk of egg consumption. These subjects, considered hyperresponders, showed significant increases ($P<0.025$) at both 4 wk and 8 wk after egg consumption in total cholesterol and LDL cholesterol levels, and at 8 wk in total cholesterol/HDL cholesterol ratio. The remaining 22 hyporesponders showed no change in any of the variables measured at 4 wk or 8 wk after egg consumption. In view of the high nutritional value of eggs, a blanket ban on eggs is not justified. However, since up to one-third of the population may be hyperresponders, knowing the response of an individual is important before making the egg a regular item of the diet.

Key words : egg dietary cholesterol cholesterol atherosclerosis

INTRODUCTION

It is now fairly well established that hypercholesterolaemia is a major risk factor for atherosclerosis (1). On the other hand, high dietary cholesterol intake has a predictable hypercholesterolaemic effect (2). Egg is a commonly consumed high-cholesterol item of the diet. On the basis of these facts, restriction of egg intake has been recommended as one of the measures for prevention and management of coronary heart disease (3). However, this recommendation ignores a few other facts. Cholesterol is a metabolic necessity, and if the dietary intake is inadequate to meet the physiological requirements, it is synthesized in the body. Further, endogenous synthesis of cholesterol is subject to homeostatic regulation such that it tends to regulate blood cholesterol levels. In view of these factors, the conflicting results of

*Corresponding Author : E-mail: rambij@hotmail.com

experimental studies on the effect of dietary eggs on blood cholesterol levels (4-7) are not difficult to explain. The results would depend on the cholesterol content and hypo- or hyper-cholesterolaemic effect of other items in the diet, may be affected by ethnicity, physical activity, hormonal status and body weight, and are also markedly affected by genetic factors (8). Since most of the previous studies on the subject have been on western populations, their results cannot be extrapolated to Indians because of the differences in dietary habits, and possibly also in relevant genetic factors. From our previous preliminary study, it was tentatively concluded that in young healthy Indian subjects on a vegetarian diet, consuming one egg per day raises serum cholesterol levels at 4 wk, but in the majority baseline values are restored by 8 wk in spite of continuing egg consumption. However, some hyperresponders were found to have elevated serum cholesterol even at 8 wk (9). The present report is based on continuation of the study on additional subjects which has permitted more firm conclusions.

METHODS

Subjects

Forty-eight healthy adult volunteers were recruited for the study in two phases. Of the volunteers enlisted, 34 (22 men, 12 women; age 25.7±5.8 years; BMI 20.8±2.3 kg/m^2) completed the study. The subjects were normolipidaemic, and none of the subjects smoked or used alcohol.

Experimental design

The study was conducted in the form of a randomised controlled trial with a cross-over design. The study started with a lead-in period of 2 wk, during which the subjects were requested to consume a relatively constant egg-free lacto-vegetarian diet, make no changes in the cooking medium, avoid baked foods (because they may contain eggs), and maintain a relatively constant physical activity.

After the lead-in period of 2 wk, the subjects were randomly divided into two groups, Group I (n=17; 12 male, 5 female; 4 lacto-vegetarians, 4 lacto-ovo-vegetarians, 9 non-vegetarian) and Group II (n=17; 10 male, 7 female; 7 lacto-vegetarians, 5 lacto-ovo-vegetarians, 5 non-vegetarian). Group I consumed for 8 wk one boiled egg every day but otherwise the diet continued to be as during the lead-in period (experimental treatment). Group II consumed for 8 wk a diet similar to that during the lead-in period (control). At the end of the 8 wk dietary period, the two groups crossed over for a period of 8 wk (Fig. 1). Fasting blood samples were collected at the beginning of the lead-in period

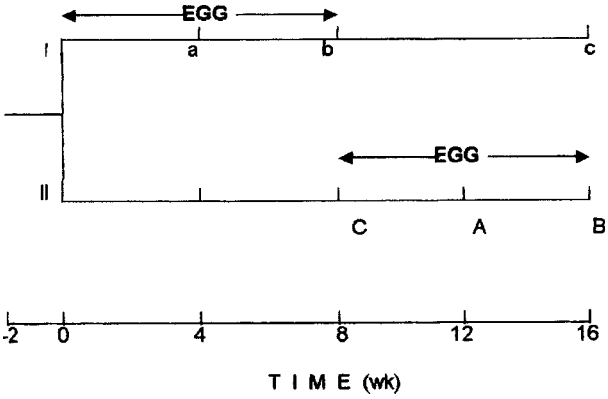

Fig. 1 : The experimental design. After confirming that there was no significant period effect, the results of the two groups were pooled. Values c and C were pooled to get the data for control (egg-free diet) treatment. Values a and A were pooled to get the 4 wk egg data, and values b and B were pooled to get the data for 8 wk of experimental (egg consumption) treatment.

(-2 wk), the end of the lead-in period (0 wk), 4 wk, 8 wk, 12 wk and 16 wk.

The protocol of the study was approved by the Ethics Committee of the All India Institute of Medical Sciences and the volunteers gave their informed written consent for participation in the study.

Diet

When required by the study, the subjects had the boiled egg at any meal of the day, usually at lunch. The eggs provided to the subjects weighed about 50 g each, and their composition was as shown in Table I. Since the cholesterol content of the eggs was 600 mg/100g, and each egg weighed about 50 g, the cholesterol intake increased by 300 mg/d during the experimental (egg) period as compared to the control period. Egg was the major source of dietary cholesterol during the experimental period, and milk and milk products its only source during the control period. The subjects maintained a report card in which they made a daily entry regarding taking, or not taking, the egg, and also recorded any major changes in their diet or physical activity.

Measurements

Serum lipid profile was assessed from the mean of values obtained on two consecutive days. The measurements included total cholesterol, total triglycerides, HDL cholesterol and LDL cholesterol, which were estimated using kits from Randox Laboratories Ltd., Ardmore, U.K. Briefly, cholesterol or triglycerides in the sample were oxidised enzymatically releasing hydrogen peroxide. Hydrogen peroxide, in the presence of peroxidase, interacts with 4-chlorophenol and 4-aminoantipyrene to yield quinoneimine, which can be measured colorimetrically from absorbance at 546 nm. HDL cholesterol was measured after precipitating LDL and VLDL by addition of phosphotungstic acid in the presence of magnesium ions. LDL was measured after precipitating it with heparin at pH 5.04. The inter-assay and within-assay variations were respectively 6.98% and 3.64% for cholesterol, 13.85% and 3.02% for triglycerides, and 11.77% and 2.74% for HDL. VLDL cholesterol and total cholesterol:HDL cholesterol ratio were calculated from the above measurements.

Statistical analysis

The values of all outcome measures at –2 wk, 0 wk, 4 wk, 8 wk, 12 wk and 16 wk were first tabulated for the two groups separately. There was no period effect on any of the parameters studied. Therefore pooling the results from the two groups was considered valid. Then the 16 wk values of Group I and 8 wk values of Group II were pooled to get the control (no egg) values. This was considered appropriate because these were the values based on antecedent egg-free diet for 8 wk in Group I and for 10 wk in Group II. The 8 wk values of Group I and 16 wk values of Group II were pooled to get the effect of the egg treatment for 8 wk (Fig. 1). The 'no egg' values were compared with 'egg values' by Student's t-test for paired observations. Similar pooling and comparisons were also done for 'no egg' treatment versus 4 wk of 'egg' treatment to see the time course of changes, if any. A comparison was also made to see the changes, if any, during the lead-in period. Since 0 wk values were compared with 4 wk as well as 8 wk values by Student's t-test, differences were considered significant if $P<0.05/2$, i.e. $P<0.025$, using Bonferroni's correction.

RESULTS

The characteristics of subjects are given in Table II. There was no significant difference between the subjects in the two groups with respect to the characteristics shown in the Table as well as in terms of the serum lipid profile. There was no significant change in body weight or body mass index (BMI) during the course of the study.

Since there was no period effect on any of the parameters studied, the results from the two groups were pooled. The lipid profile at the end of the control (egg-free) dietary period and that after 4 wk and 8 wk of experimental treatment (egg consumption) has been shown in Table III. There were no significant differences in any of the parameters either after 4 wk or after 8 wk of egg consumption except in total cholesterol/HDL cholesterol ratio which was significantly increased after 8 wk.

TABLE I: Composition of the eggs used in the study.

Constituent	*Amount*
Protein (g/100 g egg)	10.04
Fat (g/100 g egg)	11.62
Saturated fatty acids (%)	32.62
Monounsaturated fatty acids (%)	45.70
n-6 Polyunsaturated fatty acids (%)	15.56
n-3 Polyunsaturated fatty acids (%)	0.29
Cholesterol (g/100 g egg)	600.00
Energy (kcal/100 g egg)	155.78

TABLE II: Characteristics of subjects.

	Group I	*Group II*
Age (years)	26.7±7.66	24.6±3.31
Weight (kg)	59.6±9.58	54.0±6.57
Height (m)	1.66±0.10	1.64±0.06
BMI (kg/m^2)	21.35±2.28	20.03±2.14
Haemoglobin (g/100 mL)	12.5±1.81	12.2±1.78
Fasting plasma glucose (mg/100 mL)	92.5±9.91	85.4±10.22

All values are mean±S.D.

Scrutiny of individual responses revealed that twelve of the subjects had a considerable increase in serum LDL cholesterol, ranging from + 15.4% to +72.8% after 8 wk of egg consumption. Thus 12 subjects out of 34 (35.4%) had an appreciable rise in LDL cholesterol after 8 wk of egg consumption. The result of separate analysis of only this group is given in Table IV. As can be seen from the Table, there were significant increases at both 4 wk and 8 wk

TABLE III: Serum lipid profile after control and experimental treatment.

Parameter	*After control (egg-free diet) treatment*	*After experimental (egg consumption) treatment*	
		4 wk	*8 wk*
Total cholesterol	183.72±27.90	189.84±24.55	191.84±29.63
LDL cholesterol	104.73±29.81	112.34±26.20	115.42±32.47
HDL cholesterol	52.54±12.43	51.95±11.43	50.05±11.82
VLDL cholesterol	25.33±7.32	25.65±8.59	27.36±9.07
Total triglycerides	114.54±30.73	116.71±33.61	120.50±35.61
Total cholesterol/HDL ratio	3.73±1.24	3.85±1.11	4.07±1.31*

All values are Mean±S.D. All values except Total Cholesterol/HDL ratio are in mg/dL.
*P<0.025, control vs. 8 wk.

TABLE IV: Serum lipid profile after control and experimental treatment in hyperresponders.

Parameter	*After control (egg-free diet) treatment*	*After experimental (egg consumption) treatment*	
		4 wk	*8 wk*
Total cholesterol	172.42±24.73	191.18±27.89*	204.10±28.42*
LDL cholesterol	90.34±26.36	113.72±27.75*	127.89±31.77*
HDL cholesterol	54.20±13.87	51.53±11.78	47.66±12.72
VLDL cholesterol	26.85±8.67	25.60±9.90	26.03±11.46
Total triglycerides	126.19±41.58	126.06±43.76	128.04±49.08
Total cholesterol/HDL ratio	3.42±1.32	3.94±1.33	4.55±1.40*

All values are Mean±S.D. All values except Total Cholesterol/HDL ratio are in mg/dL.
*P<0.025, compared to control.

TABLE V: Serum lipid profile after control and experimental treatment in hyporesponders.

Parameter	*After control (egg-free diet) treatment*	*After experimental (egg consumption) treatment*	
		4 wk	*8 wk*
Total cholesterol	189.89±28.10	189.11±23.21	185.15±28.70
LDL cholesterol	112.58±29.41	111.59±25.96	108.62±31.47
HDL cholesterol	51.63±11.82	52.17±11.51	51.36±11.39
VLDL cholesterol	24.50±6.55	25.67±8.03	28.09±7.68
Total triglycerides	108.18±21.40	111.61±26.35	116.38±26.10
Total cholesterol/HDL ratio	3.90±1.20	3.80±1.00	3.80±1.21

All values are Mean±S.D. All values except Total Cholesterol/HDL ratio are in mg/dL.

after egg consumption in total cholesterol and LDL cholesterol levels, and at 8 wk in total cholesterol/HDL cholesterol ratio.

The results obtained on the 22 subjects whose LDL cholesterol response to egg at 8 wk was less than +15% were also analysed separately (Table V). These subjects showed no change in serum lipid profile 4 wk or 8 wk after egg consumption.

DISCUSSION

When all the 34 subjects of the present study are considered together, there is no significant change in lipoprotein profile associated with ingestion of one boiled egg per day except for a hint of an adverse effect in terms of an increase in the total cholesterol/HDL cholesterol ratio at 8 wk (Table III). However, this conclusion conceals marked individual variation, and is hence not valid. Therefore we have split the subject population depending on whether the change in LDL cholesterol as a result of egg consumption at 8 wk is greater than +15% (hyperresponders) or less than that (hyporesponders). The criterion of +15% change in LDL cholesterol had to be chosen arbitrarily because although the phenomenon of hyperrespoders is known (10), the criterion for applying the label is not established. Using our criterion, 12 subjects out of 34 (35.4%) were found to be hyperresponders. Our results are similar to some studies from the west in which also about one-third of the population consists of hyperresponders

(11). However, the number of genuine hyperresponders may be less than 12 in our study because cholesterol levels are subject to marked chance fluctuations (10). Although the cholesterolaemic response to dietary cholesterol is believed to be predominantly genetically determined (8), an interesting observation is that in the present study, the control total cholesterol and LDL cholesterol levels in hyperresponders are lower than the corresponding levels in hyporesponders (respectively, 172.42±24.73 vs. 189.89±28.10 mg/dL, $P<0.07$; and 90.34±26.36 vs. 112.58±29.14 mg/dL, $P<0.03$). However, this observation may be the result of the selection procedure itself. Subjects whose serum cholesterol, through chance fluctuations, happened to be low after control treatment increased their probability of meeting our criteria of hyperresponders. The reverse argument would also explain why the high value of serum cholesterol after control treatment in hyporesponders may not have any pathophysiological basis (10). It is possible that the cholesterolaemic response to dietary cholesterol intake is affected by the habitual dietary cholesterol intake (10), but from our limited data, no clear relationship could be deduced between dietary habits and response to egg intake.

While looking for gender differences, if any, it was observed that while there were only two women hyperresponders out of 12 female subjects, out of the 22 male subjects, 10 were hyperresponders ($P=0.09$, chi-square test). This is somewhat consistent with Herron et al's observation that in their study only the male hyperresponders showed an increase in the LDL/HDL ratio in response to cholesterol feeding whereas the female hyperresponders did not (11). Thus it seems that, on the whole, females are less susceptible to the adverse effects of cholesterol feeding.

Egg is a dietary item of considerable interest because it is a relatively inexpensive source of high quality protein, folate and other B vitamins, alpha-tocopherol and unsaturated fatty acids (12), choline, and also antioxidants such as lutein and zeaxanthin (13). But its high cholesterol content has inspired second thoughts about the confidence with which it can be recommended as a routine item of the diet. However, a blanket ban on eggs is not justified in view of the known individual variation in the cholesterolaemic response to egg. Further, the total dietary cholesterol intake of an individual depends on the overall composition of the diet. This is particularly relevant to India where the dietary habits of even the non-vegetarians are predominantly lactovegetarian or lacto-ovo-vegetarian. Therefore the major sources of dietary cholesterol in India are milk, butter and egg. Since milk and butter are taken sparingly, taking one egg containing 250 mg cholesterol per day would still leave the cholesterol intake within the permissible limit of 300 mg per day set in the National Cholesterol Education Program (NCEP) Step I diet (14). But no study before ours (9) had examined the effect of ingesting eggs on an Indian population against the background of an Indian vegetarian diet. Our study has confirmed the expectation that in such a setting in at least two-thirds of the subjects, there would be no adverse effect on the lipoprotein profile as a result of ingestion of one egg per day. However, the study has also revealed that, as in the Caucasian populations, up to one-third of the subjects show a hypercholesterolaemic response to the ingestion of egg. Therefore knowing the response of an individual to eggs is important before making the egg a regular item of the diet. In long-term studies, more than one trial with consuming an

egg for at least two weeks is essential to define the response of an individual (10), which is rather cumbersome, and hence impractical on a large scale. Therefore it would be desirable to have a convenient screening test. Whether the screening can be done by an acute cholesterol tolerance test is a subject for further studies. More studies are also needed to see whether the results of the present study can be extrapolated to older subjects, and to those at high risk for atherosclerosis. Although indications are that hyperresponders respond to dietary cholesterol by enhancing reverse cholesterol transport as indicated by higher cholesteryl ester transfer protein (CETP) and lecithin:cholesterol acyltransferase (LCAT) activity (11), further studies may throw more light on the mechanisms underlying the magnitude of the cholesterolaemic response to the dietary challenge. Finally, more studies are also needed to examine whether the level of cholesterol intake during infancy and childhood influences the development of tolerance to dietary cholesterol in adult life.

ACKNOWLEDGEMENTS

We would like to thank the Indian Council of Medical Research for financial support (ID No. 2000-00460). We are grateful to the volunteers for their cooperation. Ms. Promila Kapoor and Ms. Suman Thakur provided able technical assistance, and Mr. Satish Sachdeva provided efficient secretarial assistance.

REFERENCES

1. Grundy SM. Cholesterol and coronary heart disease: a new era. *JAMA* 1986; 256: 2849–2858.
2. Hopkins PN. Effect of dietary cholesterol on serum cholesterol: a meta-analysis and review. *Am J Clin Nutr* 1992; 55: 1060–1070.
3. Weggemans RM, Zock PL, Katan MB. Dietary cholesterol from eggs increases the ratio of total cholesterol to high-density lipoprotein cholesterol in humans: a meta-analysis. *Am J Cli Nutr* 2001; 73: 885–891.
4. Sacks FM, Salazar J, Mitter L, Foster JM, Sutherland M, Samonds KW, Albers JJ, Kass EH. Ingestion of egg raises plasma low-density lipoproteins in free-living subjects. *Lancet* 1984; 1: 647–649.
5. Sutherland WHF, Ball MJ, Walker H. The effect of increased egg consumption on plasma cholesteryl ester transfer activity in healthy subjects. *Eur J Clin Nutr* 1997; 51: 172–176.
6. Kestin M, Clifton PM, Rouse IL, Nestel PJ. Effect of dietary cholesterol in normolipidemic subjects is not modified by nature and amount of dietary fat. *Am J Clin Nutr* 1989; 50: 528–532.
7. Vorster HH, Benade AJS, Barnard HC, Locke MM. Silvis N, Venter CS, Smuts CM, Engelbrecht GP, Martha MP. Egg intake does not change plasma lipoprotein and coagulation profiles. *Am J Clin Nutr* 1992; 55: 400–410.
8. Howell WH, McNamara DJ, Tosca MA, Smith BT, Gaines JA. Plasma lipid and lipoprotein responses to dietary fat and cholesterol: a meta-analysis. *Am J Clin Nutr* 1997; 65: 1747–1764.
9. Chakrabarty G, Bijlani RL, Mahapatra SC, Mehta N, Lakshmy R, Vashisht S, Manchanda SC. The effect of ingestion of egg on serum lipid profile in healthy young free-living subjects. *Indian J Physiol Pharmacol* 2002; 46: 492–498.
10. Katan MB, Beynen AC. Characteristics of human hypo-and hyperresponders to dietary cholesterol. *Am J Epidemiol* 1987; 125: 387–399.
11. Herron KL, Vega-Lopez S, Conde K, Ramjiganesh T, Shachter NS, Fernandez ML. Men classified as hypo- or hyperresponders to dietary cholesterol feeding exhibit differences in lipoprotein metabolism. *J Nutr* 2003; 133: 1036–1042.
12. Hu FB, Manson JE, Willett WC. Types of dietary fat and risk of coronary heart disease: a critical review. *J Am Coll Nutr* 2001; 20: 5–19.
13. Handelman GJ, Nightingale ZD, Lichtenstein AH, Schaefer EJ, Blumberg JB. Lutein and zeaxanthin concentrations in plasma after dietary supplementation with egg yolk. *Am J Clin Nutr* 1999; 70: 247–251.
14. National Cholesterol Education Programme. Report of the expert panel on population strategies for blood cholesterol reduction. Executive summary. *Arch Intern Med* 1991; 151: 1071–1084.