Dietary cholesterol, eggs and coronary heart disease risk in perspective

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Summary
The idea that dietary cholesterol increases risk of coronary heart disease (CHD) by turning into blood cholesterol is compelling in much the same way that fish oil improves arthritis by lubricating our joints! Dietary cholesterol, chiefly in the form of eggs, has long been outlawed as a causative agent in CHD through its association with serum cholesterol. However, the scientific evidence to support a role for dietary cholesterol in CHD is relatively insubstantial in comparison with the incontrovertible link between its circulating blood relative in low density lipoprotein (LDL) cholesterol and CHD. Interpretation of the relationship between dietary cholesterol and CHD has been repeatedly confounded by an often inseparable relationship between dietary cholesterol and saturated fat. It has also been exaggerated by the feeding of unphysiologically high intakes of eggs. Nonetheless, numerous studies have shown that dietary cholesterol can increase serum LDL-cholesterol, but the size of this effect is highly variable between individuals and, according to over 30 years of prospective epidemiology, has no clinically significant impact on CHD risk. Variation in response to dietary cholesterol is a real phenomenon and we can now identify nutrient–gene interactions that give rise to this variation through differences in cholesterol homeostasis. More importantly, to view eggs solely in terms of the effects of their dietary cholesterol on serum cholesterol is to ignore the potential benefits of egg consumption on coronary risk factors, including obesity, diabetes and metabolic syndrome. Cardiovascular risk in these conditions is largely independent of LDL-cholesterol. These conditions are also relatively unresponsive to any LDL-cholesterol raising effects of dietary cholesterol. Treatment is focused primarily on weight loss, and it is in this respect that eggs may have a new and emerging role in facilitating weight loss through increased satiety.

Keywords: coronary heart disease (CHD), dietary cholesterol, low density lipoprotein (LDL)
Dispelling the mythology

A myth can be defined as a popular idea on a natural phenomenon. The popular idea in this context has always been that dietary cholesterol is equivalent to blood cholesterol; the natural phenomenon that raised blood cholesterol increases risk of CHD. While there is no longer debate over the role of cholesterol transported in LDL in CHD, the popular idea that dietary cholesterol increases CHD by increasing LDL prevails.

The Seven Countries Study was a landmark study in the history of nutritional medicine that founded what was to later become an unequivocal relationship between dietary saturated fat and serum LDL-cholesterol. It also led Keys and Hegsted (Hegsted et al. 1965; Keys et al. 1965) to formulate equations to predict how the relative amounts of dietary saturated and polyunsaturated fats and cholesterol in our diet influence serum cholesterol. While these equations have been reworked many times over the intervening years, the later addition and quantitative contribution of dietary cholesterol in the dietary formula has always been a contentious issue, not least between the two authors. Keys acknowledged in 1965 that while dietary cholesterol should not be ignored in advice to lower serum cholesterol, attention to this factor rather than to the nature of dietary fat would accomplish little.

The proposed relationship between dietary cholesterol and CHD is indirect and mediated through increased serum LDL-cholesterol. As eggs represent the richest source of dietary cholesterol in westernised diets, a single egg yolk containing from 50 to 250 mg of cholesterol depending on its size, they have been used exclusively as a vehicle for the delivery of dietary cholesterol in human interventions and as a marker of dietary cholesterol intake in prospective cohort studies. The endpoints from these studies have provided two important types of information: one based on changes in serum cholesterol, and thus by inference CHD risk; the other on the power of association between egg consumption and the incidence of CHD death. Since the Department of Health’s 1994 COMA report on nutritional aspects of cardiovascular disease, there has been no further reference to dietary cholesterol reduction and at present, the UK recommendations still remain at 245 mg of dietary cholesterol per day.

Does dietary cholesterol raise serum cholesterol?

A host of cross-sectional, prospective cohort and intervention studies in the 1970s and 80s fuelled debate on the potential of dietary cholesterol, chiefly in the form of eggs, to raise serum cholesterol and thus, increase CHD risk (Mann 2000). However, much of the prospective epidemiology was confounded by the fact that dietary cholesterol and saturated fat occur together in the diet so that their levels in the diet are highly correlated. It later transpired that, in many cases, the methods of dietary assessment were inadequate to accurately describe dietary intakes, and that the statistical techniques were of insufficient power to distinguish between the cholesterol-raising effects of saturated fat and cholesterol. Moreover, the majority of early human interventions were conducted against a backdrop of a high saturated fat diet (low P : S ratio), and in most cases, compared dietary extremes and unphysiologically high intakes of dietary cholesterol (>400–1000 mg/day).

Data from cholesterol-feeding in animals has been used with effect to support human studies. While it is all too easy to dismiss the findings from animal studies on the grounds that extrapolation to humans is unreliable, in this particular case, this argument may be well founded. Cholesterol-feeding in animals provides more information about the experimental toxicology of what, in dietary terms, is a foreign substance to an animal adapted to a high-carbohydrate diet. It is also difficult to ignore cross-species differences in susceptibility to coronary atherosclerosis which arise, in part, from differences in lipoprotein physiology and thus the transport of serum cholesterol in LDL and high density lipoprotein (HDL). While this is exemplified between species that are highly susceptible or resistant to dietary-induced atherosclerosis, such as the rabbit and the rat respectively, significant differences in lipoprotein physiology also exist between non-human primates and man.

Despite the practical and statistical shortcomings of early studies, a general consensus was reached that serum cholesterol increased on average by 0.01 mmol/L for every 100 mg of added dietary cholesterol (Hegsted 1986). This increase in total serum cholesterol included elevations in both LDL and the cardioprotective HDL-cholesterol; it was usually evident against a background diet that was high in saturated fat (low P : S ratio <0.5) and appeared to be highly variable between different individuals.

Some of the most scientifically rigorous studies to address the effects of dietary cholesterol on serum cholesterol and lipoproteins were to follow in the 1990s. These included two separate studies in healthy young men (Ginsberg et al. 1994) and young women consuming an American Heart Association (AHA) Step 1 diet (30% of energy as fat; 9% of energy as saturated fat) (Ginsberg et al. 1995). In the first study, 20 young men...
consumed 0, 1, 2 and 4 eggs/day in a randomised, four-way cross-over design for 8 weeks. The study examined a range of lipoprotein responses, including an assessment of postprandial response to a high-fat test meal. While there was no effect of egg-feeding on postprandial response, there was an increase in total serum cholesterol (0.038 mmol/L per 100 mg of added dietary cholesterol), that was linear for both total and LDL-cholesterol across intakes of dietary cholesterol ranging from 128 to 858 mg/day. Young women showed a greater increase in serum cholesterol in response to 0, 1 and 3 eggs/day in a three-way cross-over design for 8 weeks (0.073 mmol/L per 100 mg of added dietary cholesterol). The response was again linear for total, LDL- and HDL-cholesterol across intakes of dietary cholesterol of from 125 to 745 mg. There was a greater increase in LDL-cholesterol and less individual variation in women compared with men. In each of these studies, the background diet was believed to account for the increases in LDL-cholesterol being less than that previously reported by Keys and Hegsted (Hegsted et al. 1965; Keys et al. 1965). While the artificially high intakes of dietary cholesterol will no doubt have contributed to the statistical significance of the linear responses in LDL-cholesterol in both men and women, it is still difficult to refute the effects on LDL at lower dietary intakes and the variation in response. Similar findings were reported in hyperlipidaemic patients consuming an AHA Step 1 diet and either 2 eggs/day or a placebo for 12 weeks (Knopp et al. 1997). Hypercholesterolaemic patients showed a non-significant increase in LDL-cholesterol (+0.07 mmol/L), whereas patients with combined hyperlipidaemia (raised serum cholesterol and triglyceride) were more sensitive to dietary cholesterol (+0.3 mmol/L). Both groups showed significant increases in HDL-cholesterol. It may be concluded from these carefully conducted studies with a background diet low in total and saturated fat, that dietary cholesterol can raise total serum cholesterol by increasing both LDL- and HDL-cholesterol. This effect is evident at intakes of cholesterol of less than 400 mg/day, but the effects are small and any impact of increased LDL on CHD risk is potentially countered by increases in HDL. The effects of dietary cholesterol on the ratio of total to HDL-cholesterol, a discriminating marker of CHD risk, has been addressed in a meta-analysis (Weggemans et al. 2001). While dietary cholesterol was shown to increase this ratio, the interpretation of this result was complicated by the fact that the change in the ratio was a function of the component parts, i.e. LDL and HDL. This meant that dietary cholesterol appeared to have the most detrimental effect on LDL in subjects with the lowest total cholesterol : HDL-cholesterol ratio and thus the lowest CHD risk, a finding that was unexpected and difficult to explain.

**Does dietary cholesterol or egg consumption increase CHD risk?**

Having concluded that dietary cholesterol can increase serum cholesterol, the next most important question to ask is whether this increase translates into a clinically relevant increase in CHD risk? Over 30 years of prospective epidemiological surveys of CHD risk have consistently found no independent relationship between dietary cholesterol or egg consumption and CHD risk (McNamara 2000). A more recent study that examined data from two prospective cohort studies, the Nurses' Health Study and the Health Professionals Follow-up Study, totalling over 1 million subjects (Hu et al. 1999), could find no significant difference in cardiovascular risk between groups consuming less than one egg per day and those consuming more than one egg per day. It can therefore be concluded that dietary cholesterol, chiefly in the form of eggs, does not contribute to CHD risk. Since variation in the LDL response to dietary cholesterol may provide valuable insight into the relationship between dietary fat and CHD, and increase the potential to optimise diets for individualised nutritional health, much attention has focused on the underlying molecular basis for this phenomenon.

**Metabolic and genetic origins of variation in lipid response to dietary cholesterol**

Katan and Beynen (1983) were the first to report hyper- and hypo-responsiveness to dietary cholesterol in humans. This variation was later confirmed by the same authors in response to extreme contrasts of dietary cholesterol intakes (625–989 mg/day) in relatively small groups of subjects (Beynen & Katan 1985; Katan et al. 1986). Further study of this phenomenon in a much larger group of normo- and hyper-lipidaemic subjects consuming a low saturated fat, high-fibre diet and physiologically relevant intakes of dietary cholesterol [first 2 and then 7 eggs per week (100–300 mg/day) for 4 months] could find no significant difference in serum cholesterol between the two treatments, but did identify a subgroup of 58 individuals with a mean increase in LDL-cholesterol of more than 5% (Edington et al. 1987). In a second phase of this experiment, this subgroup was re-examined on higher intakes of dietary cholesterol (no eggs and then 9 eggs per week: 100–400 mg of cholesterol/day) for a further 3 months, but once...
again, showed no significant increase in LDL-cholesterol (Edington et al. 1989). It was concluded that physiological intakes of dietary cholesterol in combination with a standard cholesterol-lowering diet, produce only small effects on serum cholesterol that are of negligible clinical relevance. There was also no consistent evidence of hyper- and hypo-responsiveness to dietary cholesterol as had been previously shown under more extreme dietary conditions. Nevertheless, around 20–30% of subjects studied are commonly reported to show increased responsiveness to dietary cholesterol. This finding is not unexpected, especially in view of our increased understanding of how nutrient–gene interactions influence cholesterol homeostasis.

**Effects of dietary cholesterol on cholesterol homeostasis**

The discovery, by Goldstein and Brown (1986), of the mechanism by which cells regulate their content of cholesterol and thus, inadvertently, regulate blood cholesterol levels via the LDL-receptor pathway, revolutionised our understanding of cholesterol homeostasis. It also unfortunately spawned a number of extreme human and animal studies designed to test how dietary constituents, including eggs, increase LDL-cholesterol by suppressing the activity of this pathway.

Cells regulate their intracellular pool of cholesterol by switching on the production of LDL receptors that migrate to the surface of cells and can then extract cholesterol from the blood in the form of LDL, and/or by increasing their own production of cholesterol. The cholesterol content in cells is also significantly influenced by the amounts of dietary and biliary cholesterol that are actively reabsorbed and excreted back into the intestinal lumen by an interplay between cholesterol transporters and cholesterol efflux proteins. Cells meet their requirements for cholesterol by adjusting the uptake of serum LDL-cholesterol and cholesterol absorption from the intestine or by increasing cholesterol biosynthesis. Cholesterol absorption and the uptake of LDL are inversely related to rates of cholesterol biosynthesis so that cells receiving increased delivery of cholesterol from these two sources synthesise less of their own cholesterol. Both the receptor-mediated uptake of LDL-cholesterol and cholesterol absorption are also highly sensitive to changes in dietary fat, notably saturated fat, and dietary cholesterol. As a good example of a nutrient–gene interaction, high intakes of dietary cholesterol can downregulate the LDL-receptor gene, suppress the production of LDL receptors and in doing so, raise LDL-cholesterol. However, over a physiological range of dietary cholesterol intakes, the body compensates for this effect by decreasing the amount of dietary cholesterol that is absorbed in the intestine (Ostlund et al. 1999). Variation in the sensitivity of this compensatory reduction in cholesterol absorption is believed to account for much of the inter-individual variation seen in response to increased dietary cholesterol. Likewise, variation in the sensitivity of the LDL-receptor pathway is potentially another source of variance in response to dietary cholesterol and, more importantly, saturated fat. The best working examples of how these pathways operate are provided by the actions of LDL-lowering drugs such as the statins and blockers of cholesterol absorption such as ezetamibe, both of which result in an up-regulation of LDL-receptor activity but by inhibiting cholesterol biosynthesis and cholesterol absorption, respectively. Dietary-induced changes that are analogous to the effects of statins and cholesterol absorption blocking drugs would be to substitute dietary saturated fats with poly- or mono-unsaturated fats and to impair cholesterol absorption in the intestine with plant sterols, respectively.

**Influence of genetic variation on the responsiveness to dietary cholesterol**

There are a number of common polymorphic genes that code for key regulatory proteins in cholesterol metabolism that may explain variation in inter-individual response to dietary cholesterol. The most notable of these is the gene for apoprotein E (Apo E), a protein found on the surface of lipoproteins that facilitates the binding and uptake of cholesterol-carrying lipoproteins. Apo E exists in different protein isoforms (E4, E3, E2) that differ in their amino acid composition and which express variable affinity for the LDL and remnant receptors in the order of E4 > E3 > E2. As apo E4 binds with higher affinity to these receptors, it effectively over-delivers cholesterol to cells. In theory, this produces a relative suppression of the production of LDL receptors, resulting in higher serum LDL-cholesterol in individuals expressing apo E4 (approximately 10–15% of the UK population). In addition, cholesterol absorption is up-regulated in the intestine so that apo E4 carriers tend to be high absorbers of dietary and biliary cholesterol, but have lower rates of endogenous cholesterol synthesis in cells (Kesaniemi et al. 1987). As a direct consequence, carriage of an apo E4 allele tends to increase responsiveness to changes in dietary cholesterol (Sarkinen et al. 1998). Another potential source of genetic variation that may contribute to variation in response to dietary cholesterol through differences in cholesterol absorption.
is via polymorphisms in proteins in the intestine known as ATP-binding cassette (ABC) cholesterol efflux proteins. Up-regulation of cholesterol excretion back into the intestine via these efflux proteins forms an important part of the mechanism that compensates for increased dietary cholesterol. Mice in which the gene for this protein has been deleted (ABC-1 knock-out mice) lose the ability to compensate for increased dietary cholesterol and as a result, show significant increases in serum cholesterol compared with controls (McNeish et al. 2000). Similarly, in humans, a common genetic polymorphism coding for an efflux protein known as ABCG5-gene (G/G variant) has been associated with reduced cholesterol excretion and increased responsiveness to dietary cholesterol (Weggemans et al. 2002). The LDL-lowering effects of plant sterols may also be partly explained by a decrease in cholesterol absorption produced in response to an increase in the expression of ABC-AI cholesterol efflux protein (Plat & Mensink 2002).

**Dietary cholesterol and eggs in perspective of other coronary risk factors**

Another possible explanation for why dietary cholesterol cannot be independently associated with CHD is that the relationship is mediated through a single risk factor, LDL-cholesterol. Multiple layers of evidence exist to support the relationship between a raised LDL-cholesterol and CHD, but dietary-induced changes in LDL-cholesterol have never provided an adequate explanation for the relationship between diet and CHD, perhaps because LDL-cholesterol has limited power to discriminate CHD risk within populations. It should be appreciated that LDL is just one risk factor of many that contribute to the multi-factorial aetiology of cardiovascular disease. A much more prevalent source of attributable cardiovascular risk in the UK population comes from the heterogeneous clustering of metabolic complications associated with obesity, diabetes and the metabolic syndrome (Kahn et al. 2005). The pro-atherogenic features, including dyslipidaemia (raised triglycerides, low HDL), hypertension, hyperglycaemia and pro-inflammatory and pro-thrombotic tendencies, arise from insulin resistance and in combination exert multiplicative effects on coronary risk that are largely independent of LDL-cholesterol. Dietary strategies to treat this risk focus on the treatment of the individual. Since fat is the most energy-dense macronutrient, its removal is fundamental to this approach. However, there is now evidence to suggest that dietary cholesterol and eggs have a significantly reduced impact on serum LDL-cholesterol in insulin-resistant states, and may even facilitate weight loss through mechanisms of satiety.

Beynen and Katan (1983) found greater LDL-lowering in response to the removal of dietary cholesterol (eggs) in individuals with lower body mass index and high HDL-cholesterol. Reaven studied the effects of egg-feeding in 31 women with insulin resistance compared with 34 insulin-sensitive women (Reaven et al. 2001). The expectation at the outset of this study was that the insulin-resistant group would be more responsive to dietary cholesterol, but no significant effect on LDL-cholesterol could be shown, irrespective of insulin sensitivity, over a wide range of dietary intakes (up to 800 mg/day) (Reaven et al. 2001). Knopp et al. (2003) re-examined the interrelationships between insulin resistance, obesity and egg-feeding and found that while insulin resistance was associated with a moderately raised LDL-cholesterol at baseline, this must be for some inherent metabolic reason, as this group showed significantly lower increases in LDL-cholesterol in response to egg-feeding compared with the insulin-sensitive subjects, irrespective of obesity status.

Insulin-resistant and obese subjects have been shown to express higher rates of cholesterol biosynthesis and lower levels of cholesterol absorption in comparison with lean, insulin-sensitive subjects (Berglund & Hyson 2003). In theory, these effects could arise as a consequence of the failure of insulin to stimulate the LDL-receptor pathway followed by a knock-on effect of increased cholesterol synthesis, which would be to inhibit cholesterol absorption in the intestine. Whatever the mechanism, the diminished effect of dietary cholesterol from eggs on serum LDL in this group has implications for the emerging role of eggs in the promotion of weight loss in this group.

**Potential role of eggs in promoting weight loss**

Weight-loss programmes, like many of the willing participants, come in all shapes and sizes. One of the most popular practices is that of a low-carbohydrate, high-fat diet that invariably leads to a raised intake of dietary cholesterol through the increased consumption of eggs and meat. While the long-term safety of these diets, that exclude important food groups, is still questionable, they are without doubt successful in promoting weight loss at least in the short-term. They also exert either no effect or potentially favourable effects on serum LDL-cholesterol and triglycerides (Foster et al. 2003; Samaha et al. 2003). The explanation for the weight loss lies in the fact that the dieters achieve an energy deficit by simply eating
less. This is most likely to occur through a combination of reduced food choice and increased satiety. Dietary protein has long been identified as the macronutrient most closely associated with satiety. Eggs, as a rich source of high-quality protein that is low in energy, fulfill this criterion very well. Eggs have been shown to have a 50% greater satiety index as compared with ready-to-eat breakfast cereals and white bread (Holt et al. 1995), and have been shown to have an overall higher satiety value than common breakfast foods in non-obese subjects (Holt et al. 2001). While the short- and longer-term efficacy of eggs in promoting weight loss has yet to be established, evidence is beginning to emerge that they may offer a viable means of sustaining an energy deficit in overweight and obese subjects (Vander Wal et al. 2005). While it is certainly not the intention here to advocate the use of extreme diets to promote weight loss, eggs are a nutritious food that would seem appropriate for assisting weight loss in an exponentially growing proportion of the free-living population at increased CHD risk (Herron & Fernandez 2004).

Conclusion

Dietary cholesterol has been shown to produce small increases in serum LDL-cholesterol, but it should be stressed that this effect does not translate into increased CHD risk. The prevention of CHD continues to be a major public health issue in the UK population, but preventative strategies must focus on obesity, diabetes and metabolic syndrome as the most prevalent sources of increased risk. In view of the overriding importance of weight loss in reducing risk from these conditions, and evidence to suggest that people with insulin resistance are unresponsive to dietary cholesterol, the moderate consumption of eggs (1–2 eggs per day) should be actively encouraged as part of an energy-restricted, weight-losing dietary regimen. There will always be some degree of public confusion about the difference between the cholesterol that we eat and that which circulates in our blood. Take for example, the body builder eating ten eggs a day who claimed to have no fear of dietary cholesterol because he only ever eats high-density cholesterol. Unfortunately, things are not always that simple!

References

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