11. Clinical Nutrition

INDIVIDUALITY OF THE RESPONSE OF SERUM CHOLESTEROL TO DIET

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Introduction

In 1926 Mjassnikow, and in 1933 Okey & Stewart, reported that the mean serum cholesterol concentration in human subjects increased somewhat on cholesterol-supplemented diets, but that there was a considerable variability in individual response. A similar inter-individual variation in cholesterolemic response was seen in most experimental studies that followed, and the concept of hyper- and hypo-responders to dietary cholesterol became widely accepted (Connor & Connor, 1972; Reiser, 1978). However, in the numerous studies in which the effect of dietary cholesterol on serum cholesterol in humans was assessed (McGill, 1979), the response to the dietary challenge in a given subject was usually measured in one study only.

The serum cholesterol concentration in any particular individual fluctuates with a coefficient of variation of 5 to 10 per cent around the mean value (Demacker *et al.*, 1985; Keys, 1967). These fluctuations are independent of the diet and are of the same order of magnitude as the usual response to dietary cholesterol loads, which rarely exceeds 20 per cent (McGill, 1979) even with extreme dietary loads. As a result, individual responses cannot be measured precisely enough to allow classification of subjects as hypo- or hyper-responsive. Table 1 illustrates this. Six volunteers first abstained from cholesterol-rich products for 10 days, and then took six egg yolks per day for another 10 days. The study was repeated with the same subjects one year later. The average response for the group was fairly similar from one experiment to another, but the "hyper-responders" in the first experiment were not necessarily hyper-responders in the second experiment, and neither were those initially classified as hypo-responders consistently unresponsive the second time. Similar experiments had already been performed in 1942 by Messinger and coworkers (1950).

Hypo- and hyper-responders to dietary cholesterol

Although the existence of human hypo- and hyper-responders has been frequently assumed (Connor & Connor, 1972; Reiser, 1978), it has proved very difficult to substantiate this experimentally. We have carried out three controlled dietary trials with the same subjects to address the question whether individuals exist who have a consistently high or low serum cholesterol response to dietary cholesterol (Katan *et al.*, 1986). In each trial the volunteers successively consumed a low- and a high-cholesterol diet, the cholesterol component of the diets (provided by egg yolk) being the only variable. Subgroups of putative hypo- and hyper-responding subjects, with mean serum cholesterol increases of 0 and 19 per cent respectively, were selected from a larger population is a first trial and then underwent a second and third experiment. Although the response in each subject was only partly reproducible, the selected hyper-responders showed significantly higher serum cholesterol

Table 1. *Changes in serum cholesterol levels in six human volunteers after daily consumption of six egg yolks for 10 days.* Twelve months elapsed between Expt 1 and Expt 2; the design was otherwise identical. The pre-experimental and experimental serum cholesterol values were both based on two blood samples obtained on successive days. After Katan & Beynen (1983).

	Subjects					
	A	*B*	*C*	*D*	*E*	*F*
	(% change in serum cholesterol)					
Expt 1	+ 5	- 3	+17	+17	+27	+ 5
Expt 2	+16	+12	+26	+25	+ 4	+ 3

Table 2. *Effect of egg-yolk cholesterol on serum cholesterol in three controlled trials with the same subjects.* Results are expressed as means ± SD.

	Change in serum cholesterol (mmol/l)	
	Hypo-responders (n=15)	*Hyper-responders (n=17)*
Selection trial	-0.01 ± 0.21	+0.96 ± 0.27
First reproducibility trial	+0.06 ± 0.35	+0.28 ± 0.38*
Second reproducibility trial	+0.47 ± 0.26	+0.82 ± 0.35**

Change significantly different from that in the hypo-responders (one-tailed Student's *t* test): *, $P < 0.05$; **, $P < 0.005$. Based on Katan *et al.* (1986).

Table 3. *Mean serum cholesterol concentrations and cholesterol absorption in monkeys on low- and high-cholesterol diets.* Based on data taken from Lofland *et al.* (1972), Eggen (1976) and St. Clair *et al.* (1981).

	Low-cholesterol diet		*High-cholesterol diet*	
	Serum cholesterol (mmol/l)	*Cholesterol absorption (%)*	*Serum cholesterol (mmol/l)*	*Cholesterol absorption (%)*
Squirrel monkeys				
Hypo (*n*=3)	4.5		4.9	55
Hyper (*n*=4)	6.1		8.3	62
Rhesus monkeys				
Hypo (*n*=5)	2.4	46	6.6	45
Hyper (*n*=5)	3.5	60	18.1	53
African green monkeys				
Hypo (*n*=3)	3.6		3.9	37
Hyper (*n*=9)	3.6		7.3	56

responses in the second and third trial than the hypo-responders (Table 2). Standardized regression coefficients for individual responses in two experiments ranged from 0.34 to 0.53 (n=32) (Katan *et al.*, 1986).

Under less controlled conditions we found similar results. Bronsgeest-Schoute *et al.* (1979) studied the serum cholesterol response to cessation of egg consumption in subjects who habitually consumed at least one egg per day. When eggs were eliminated from the diet, daily cholesterol intake decreased from about 800 to 300 mg. Mean serum cholesterol fell only slightly (by 3%), but the individual responses varied from -20% to +8%. In 1982, 34 of these subjects were re-investigated (Beynen & Katan, 1985a), and at our request they again eliminated eggs and egg-containing products from their diet. The differences in serum cholesterol response between individuals were partly reproducible; the individual responses in 1976 and 1982 were positively correlated (r=0.32, n=34, $P<0.05$).

Thus it appears that at least part of the cholesterolemic response to dietary cholesterol in man is individually determined. It is also clear that one will always find subjects who appear hyper-responsive in one experiment and hypo-responsive in another. This is caused by the diet-independent within-person variability of serum cholesterol. In our controlled studies (Katan *et al.*, 1986) we calculated that the within-person error variance was still responsible for about 25% of the apparent variance in response between subjects even if we used 12 independent blood samples to determine each person's response to dietary cholesterol. Thus it is probably fallacious to characterize a patient as hyper- or hypo-responsive to diet therapy if this is based on the results of a few blood samples only. A large number of serum cholesterol measurements are needed before and after the dietary challenge, and even then the observed responses should be interpreted with caution.

Hypo- and hyper-responders to saturated fatty acids

In man the nature of fat in the diet is more important as a determinant of the serum cholesterol concentration than the amount of cholesterol. Thus it is relevant to know whether hypo- and hyper-responders to dietary fatty acid composition also exist, and whether hyper-responders to dietary cholesterol are also hyper-responsive to saturated fatty acids. Such information may also provide clues to the mechanisms underlying the inter-individual variation in the cholesterolemic response to diet.

We have addressed the question whether human subjects hypo- or hyper-responsive to dietary cholesterol are also hypo- or hyper-responsive to saturated fatty acids in the diet. Twenty-three subjects who participated in the three controlled trials on the effect of dietary cholesterol (Katan *et al.*, 1986) were also tested for their response to saturated *versus* polyunsaturated fatty acids. In this experiment cholesterol intake was kept constant at an average of 41 mg/MJ (almost 500 mg/day), but the energy percentage of dietary polyunsaturated fatty acids was kept at 21 per cent for the first 3 weeks and then changed to 5 per cent for the next 3 weeks; the polyunsaturated : saturated fatty acids ratios were 1.91 and 0.22 respectively. The response of serum cholesterol to the change in dietary fatty acid composition in this experiment was positively correlated with the mean response to dietary cholesterol in the three preceding experiments (r=0.50; n=23; $P<0.05$). This indicates that in humans, hyper-responsiveness to dietary cholesterol is associated with hyper-responsiveness to saturated fat.

Underlying mechanisms of aberrant response

On the basis of published work we proposed earlier that hypo-responders suppress cholesterol synthesis more effectively after cholesterol feeding than hyper-responders (Beynen & Katan, 1985b; Beynen *et al.*, 1985). Thus hyper-responders do not compensate for increased amounts of dietary cholesterol, and there is eventually an increase in serum cholesterol. We feel at present that individual variation in the efficiency of gastro-intestinal absorption of cholesterol could be the primary determinant of hypo- and hyper-responsiveness. This idea is based on animal studies with different species of monkeys. In

three studies hyper-responsive monkeys absorbed a significantly higher percentage of dietary cholesterol than did their hypo-responsive counterparts (Table 3).

Hyper-responsiveness to dietary cholesterol is associated with responsiveness to dietary saturated fatty acids, pointing to a common metabolic pathway. However, it is not yet possible to illustrate this in molecular terms. The increase in serum cholesterol after cholesterol feeding in humans is due mostly to an increase in low density lipoprotein (LDL) cholesterol concentration. Why does dietary cholesterol cause an increase in LDL cholesterol in hyper-responders, but less or not at all in hypo-responders? When cholesterol consumption is increased, more dietary cholesterol on chylomicron remnants could enter the liver in hyper- as compared to hypo-responders. The liver seems very efficient at removing chylomicron remnants from the plasma, but it responds to excessive uptake of cholesterol in at least two ways: 1) suppression of endogenous cholesterol synthesis, and 2) increased secretion of cholesterol into the blood. Dietary cholesterol does not stimulate fecal bile acid excretion in humans (Beynan & Katan, 1985b).

Regardless of the possibility (Beynen & Katan, 1985b; Beynen *et al.*, 1985) that there is a lack of inhibition of cholesterol synthesis in hyper-responders, there could also be increased hepatic secretion of cholesterol in these individuals, due to an enhanced influx of chylomicron-remnant cholesterol. The increased output of cholesterol by the liver would explain the observed increase in LDL production after cholesterol feeding (Packard *et al.*, 1983). LDL may be secreted directly into the plasma from the liver, but as a rule the very-low density (VLDL) and intermediate density (IDL) lipoprotein precursors appear first. Nestel & Billington (1983) have shown that in man cholesterol feeding caused an increase in IDL-apo B production, and that this increase was directly correlated with the rise in serum cholesterol. Thus, hyper-responders may have increased rates of LDL cholesterol production after cholesterol feeding, and this may explain their elevated plasma concentrations of LDL cholesterol. Subsequently, the number of LDL receptors, which may already be decreased in hyper-responders (Mistry *et al.*, 1981), will decrease further through down regulation (Brown & Goldstein, 1984), as shown in blood mononuclear cells (Mistry *et al.*, 1981). As a result the receptor-mediated fractional clearance of LDL decreases (Packard *et al.*, 1983), but the absolute amount of LDL cholesterol delivered to the cells by the receptor pathway increases somewhat because the concentration of substrate (LDL) is increased (Packard *et al.*, 1983). The rise in LDL production will also increase LDL clearance by the receptor-independent scavenger pathway. In this way a new equilibrium is reached in which LDL production again equals LDL catabolism.

Acknowledgements—Our studies were supported by the Netherlands Heart Foundation. We thank Ineke Zaalmink for typing the manuscript.

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