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Authors(s)	Gormley, T. R. (Thomas Ronan), Kevany, J., O'Donnell, B., et al.
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EFFECT OF PEAS ON SERUM CHOLESTEROL LEVELS IN HUMANS

T. R. Gormley

An Foras Taluntais, Kinsealy Research Centre, Malahide Road, Dublin 5

J. Kevany

Department of Community Health, Trinity College, Dublin 2.

B. O'Donnell

Eastern Health Board, River House, Chancery Street, Dublin 7

R. McFarlane

Department of Medicine & Therapeutics, University College, Dublin 4.

ABSTRACT

Fifty male and six female free living volunteers were paired in two groups based on similar total cholesterol levels. Each individual in one group ate 30 g of freeze dried peas daily over the period February 8 to March 22 while the corresponding individuals in the control group ate an isocaloric quantity of cornflakes daily.

Serum cholesterol levels and high density lipoprotein fractions (HDL cholesterol) were measured at two-weekly intervals during the experiment and the pea group had consistently lower total cholesterol values (significantly different on two of the testing dates) than the cornflake group. HDL cholesterol values were consistently higher for the pea group and the effect was significant at the six-week testing date. The effects on cholesterol and HDL cholesterol were still significant two weeks after cessation of the supplement but not after seven weeks.

The results of this experiment could have application in the context of long term modification of dietary patterns in relation to heart disease risk both in a general community sense and also in hyperlipidaemic subjects.

INTRODUCTION

A research programme was commenced at Kinsealy Research Centre in 1976 to emphasise foods of plant origin which have a hypocholesterolaemic (cholesterol lowering) effect when consumed in normal dietary quantities. It was considered that in the context of long term modification of dietary patterns in relation to heart disease risk, an educational programme emphasising a positive approach by increasing intake of foods of plant origin might be more successful than traditional negative approaches recommending decreased intake of fat. The first component of this programme was completed in 1977 when two apples per day, in addition to the normal diet, had a hypocholesterolaemic effect in free-living male volunteers (1). The hypocholesterolaemic effect of porridge was studied as the second component of the programme and the experiment was carried out over a six week period in 1978 with matched pairs of volunteers. No cholesterol lowering effect was observed nor was there any increase in HDL cholesterol (2).

It was decided to test peas as the third component of the programme in view of the hypocholesterolaemic effect of legumes — mostly dried beans — reported by a number of workers (3-6). In addition, peas are the most important horticultural field crop in Ireland and they are consumed extensively in a number of processed forms. The experiment was carried out for six weeks in the period February-March 1979 with matched pairs of volunteers. One group was obliged to consume peas and the other cornflakes in addition to their normal diet.

EXPERIMENTAL

Male and female volunteers from three Research Centres and the Administrative Centre of the Agricultural Institute in Dublin participated in the study. Their weight, height, smoking habits and approximate intake of dietary fibre were measured at the start of the experiment and their weight was recorded again at the end of the supplement period of the experiment. Approximate dietary fibre intake of the subjects was obtained based on their weekly intake of fruits, vegetables and cereal products as noted on a detailed questionnaire. A body mass index ($\frac{Wt}{Ht^2}$) was used in this test as an indicator of general nutritional status and build (7). Wt is expressed in grams and Ht in cm.

Experimental design

Fifty male volunteers and 6 females were obtained for the study giving a total of 28 pairs — males being paired with males and females with females. Volunteers were in the age group 30-50 years except for 9 who were less than 30 and 4 who were over 50 years.

Four of the volunteers smoked cigarettes, 3 smoked cigars and 2 were pipe smokers. Smokers and non-smokers were distributed between the two groups.

Preliminary blood samples were taken on January 25 and February 1 and volunteers were paired on the basis of similar total cholesterol levels into two groups. The decision as to which person in each pair would be on the pea or cornflake supplement was taken in a random fashion.

The intake of peas and cornflakes commenced on February 8 and further blood samples were taken on February 22, March 7 and March 22 i.e. 2, 4 and 6 weeks after the commencement of the supplement. The supplement was terminated on March 22 and further blood samples were taken 2 and 7 weeks later i.e. April 5 and May 10.

Intake of peas and cornflakes

One group consumed tender young freeze dried peas, (Erin Instant Brand) 30g per day, and the other 20g of cornflakes. The freeze dried peas had a pleasant crunchy texture and the volunteers were given the option of consuming them in the freeze dried form or as reconstituted peas. No particular time was specified for consumption of the peas but the volunteers were asked to fill out a log sheet stating the amount consumed, whether consumed in the freeze dried or reconstituted form, and also the time of the day when they were consumed. The group consuming cornflakes was requested to eat them at breakfast time and was allowed to use milk and sugar as required. No other dietary control was exerted in the experiment.

Analytical procedures

Blood samples were allowed to clot and the serum was separated after 4 hr. Serum was stored at 4°C and analysed within 24 hr for total cholesterol and HDL cholesterol. In order to obtain HDL cholesterol, low and very low density lipoproteins were precipitated from the sera by the method of Burnstein *et al* (8). Cholesterol was measured in the sera (total cholesterol) and in precipitated sera (HDL cholesterol) by a cholesterol esterase/cholesterol oxidase/colour reaction (9) using a kit supplied by Boehringer, Mannheim, West Germany. HDL cholesterol is expressed as a percentage of total cholesterol. The analyses were performed in duplicate and were blind in that each sample was allotted a different number on each occasion and laboratory identification was impossible until the study was completed. The blood samples taken after 4 weeks on the supplement i.e. March 7 were also tested for serum magnesium content (10) to see if the group on peas had a lower mean blood magnesium content than those on cornflakes.

The freeze dried peas were tested for dietary fibre (DF) content and also for level of pectin. The DF was estimated on 100 mg of defatted material. The starch in the material was gelatinised and the product was treated with amyloglucosidase overnight at 37°C. Four volumes of absolute ethanol were added and the resulting precipitate was collected after centrifugation and washed a number of times with 80% aqueous ethanol and also with acetone. The protein and ash content of the residue was determined on a separate sample and was subtracted to give a figure for DF. The pectin content of the freeze dried peas was determined using the procedure of Warren & Woodman (11).

RESULTS AND DISCUSSION

The daily DF intake of the peas and cornflakes groups was similar at the start of the experiment with mean daily values of 20g and 18g respectively. The mean daily intake of peas was 92% of target and cornflakes was 96% according to log records which indicated that the participants were consuming the required amounts. In the pea group 11 of the volunteers consumed peas with their dinner while 4 consumed them with their dinner or at another time. The remainder consumed the peas during the day at non-meal times. Sixteen of the volunteers on peas consumed them in the re-constituted form, 3 consumed them in the re-constituted form or in the freeze dried form on occasions, while the remainder ate the peas as a snack food in the freeze dried form.

Three of the volunteers on the peas did not finish the experiment as they found it too difficult to take the quantity specified. Of the remaining 25 volunteers on peas who did complete the experiment, 7 complained of symptoms such as slight stomach pains, a slight laxative effect, slight bloating and flatulence; 5 complained that the quantity was extra large while the remainder had no adverse comments. It should be noted that the adverse effects described above mostly occurred in the first week or two of the experiment. The amount of peas consumed represents about one and a half to twice the normal serving. (The 30g pack of freeze dried peas re-constituted to a weight of about 120g after treatment with boiling water for 2 min).

Serum cholesterol levels

The results (Tables 1 and 2) show that there were significant differences between % HDL cholesterol levels and between total cholesterol levels for groups on peas or cornflakes. The results for HDL cholesterol were not significantly different until the sixth week of the experiment when the pea group had the higher value. However, the values for the pea group were higher at the previous two testing dates but the effect was not statistically significant. There was also a carry-over effect after the cessation of the supplement as the HDL cholesterol level of the pea group was still significantly higher two weeks after cessation of the supplement (Table 1). However, after 7 weeks post-supplement the HDL cholesterol levels of the two groups were identical. Percentage HDL cholesterol levels were highest for both groups at the 7 week post-supplement stage (Table 1). This was largely due to the fact that total cholesterol levels had fallen considerably at this time due to the seasonal effect resulting in a higher percentage figure.

Table 1: Group values for % HDL cholesterol

	Pea group	Cornflake group	"t" test	SE
Preliminary (25 Jan. + 1 Feb.)	23.1	22.6	NS	± 0.64
2 weeks (22 Feb.)	24.5	23.0	NS	± 0.87
4 weeks (7 Mar.)	21.4	20.1	NS	± 0.86
6 weeks (22 Mar.)	24.4	21.5	**	± 0.64
----- Cessation of Supplement -----				
2 weeks post diet (5 Apr.)	22.1	19.9	*	± 0.92
7 weeks post diet (10 May)	26.8	26.9	NS	± 1.25

Patterns for total cholesterol showed the pea group with lower values throughout the experiment. Values were significantly lower at the 2 and 6 week testing dates and also two weeks after the supplements finished indicating that peas were having a hypocholesterolaemic effect. The two groups had a very similar cholesterol level 7 weeks after cessation of the supplement (Table 2). The cholesterol values for both groups were much lower at the last testing date. However, this is likely to be a seasonal effect as cholesterol levels tend to fall in the summer months. It could be argued that the increase in total cholesterol levels for the group on cornflakes during the experiment could be an effect of the cornflake supplement. However, this is highly unlikely as cornflakes would not be expected to raise cholesterol, and it has been shown in a previous experiment that cornflakes had no effect on total cholesterol values (2).

Table 2: Group values for total cholesterol (mg/100 ml serum)

Time	Pea group	Cornflakes group	"t" test	SE
Preliminary (25 Jan. + 1 Feb.)	235	235	NS	± 1.03
2 weeks (22 Feb.)	227	239	*	± 4.04
4 weeks (7 Mar.)	232	239	NS	± 4.02
5 weeks (22 Mar.)	231	247	**	± 3.56
----- Cessation Supplement -----				
2 weeks post diet (5 Apr.)	232	245	*	± 4.70
7 weeks post diet (10 May)	212	214	NS	± 6.12

The hypocholesterolaemic effect of peas and the positive effect on HDL cholesterol found in this experiment while not large represent an improvement in cholesterol status and therefore warrant the testing of a wide range of fruit and vegetables to see if they have a similar effect. These data agree with results found in other experiments with apples (1) and legumes (4, 6, 12, 13). Most of the legumes tested were dried beans but quantities were much larger (100-150 g dried) than the amount of peas used in this experiment.

The results raise important issues in relation to overall diet and indicate the possible significance of a high intake of vegetables and other foods of plant origin as a factor in balancing intake of foods of animal origin (14). The effects in this experiment were obtained in the absence of any dietary control other than the intake of peas and cornflakes. The amount of peas required to be consumed daily was not excessive and the peas used were not a "special preparation" in that they were a normal commercial freeze dried product of high quality (tender young peas) on sale at retail outlets in Ireland. They were packed in 30g packs which represents the required daily intake. The positive effect on HDL cholesterol is particularly significant in view of the importance being attached to this aspect of cholesterol in terms of a protective factor (15, 16). There is no reason to suppose that a similar result would not be obtained with frozen or canned peas. The data were examined to see if the time of the day when the peas were consumed and also the form, i.e. freeze dried versus reconstituted was having an effect on cholesterol values, but no pattern was observed.

The carry-over effect (Tables 1 and 2) on cholesterol reduction in the pea group which was found 2 weeks after cessation of the supplement is an important effect in that it suggests that peas might have a persistent hypocholesterolaemic effect if consumed every

second or third day rather than every day which would be more acceptable on a population basis. The point in time when the carry-over effect ceased was not determined exactly but it had gone 7 weeks after cessation of the supplement as shown by very similar group values for HDL cholesterol and also total cholesterol (Tables 1 and 2).

The mechanism by which the peas exert their hypocholesterolaemic effect is not clear. Hellendoorn (6) points out that the consumption of beans results in a more rapid bowel transit time and a greater excretion of bile acids. The faster transit time may be due to the formation of volatile fatty acids by fermentation in the intestine which stimulate the peristaltic action of the intestine. The hypocholesterolaemic action may also be partly physical in nature, viz. by absorption of bile acids onto faecal fibre (13). The DF content of the 30g of freeze dried peas consumed daily in this experiment was about 5.3g which in effect raised the daily DF intake of the pea group by about 25% i.e. from 20 to 25g of DF per person per day. Pectin has also been associated with cholesterol reduction (17-20). The pectin content of the 30g of freeze dried peas consumed daily was about 1.85g, which is unlikely to be enough by itself to decrease serum cholesterol if one holds with the dose response work of Palmer and Dixon (17). If, however, the peas were re-constituted before consumption, the pectin content was lower at 1.58g presumably due to leaching.

Potter *et al* (21) have suggested that dietary saponins can exert a hypocholesterolaemic action and postulated such an effect for the hypocholesterolaemic action of soya protein. Tests for the saponin content of the freeze dried peas were not carried out but there are reports in the literature on the presence of saponins in peas (22).

It may be possible, therefore, that the hypocholesterolaemic action of the freeze dried peas may be due in part to their content of DF, pectin, or saponins, or a combination of all three. Sirtori *et al* (23) have suggested that vegetable protein could also exert a hypocholesterolaemic effect.

Serum magnesium levels

Tests were carried out on serum magnesium levels to study if the level of pea intake was having a scavenging effect on magnesium and possibly other cations (using magnesium level as an index). Results indicate no difference in serum magnesium levels for the pea and cornflakes groups with values of 0.88 and 0.89 mmol/l respectively. This indicates little if any scavenging effect. Recent work by James *et al* (24) suggests that calcium binding (and presumably other cations) is not the problem previously thought and they have shown that while uronic acids in carrots bind calcium, they release it again in the colon when they are fermented and the calcium is absorbed there.

Weight data

The data (Table 3) show that there was no change in mean body mass (weight/height index) for the groups on peas or cornflakes during the course of the experiment indicating that overall dietary patterns did not change much. The mean weight/height index of the cornflake group was greater than for the pea group but the effect was not significant and was largely caused by two people in the cornflake group who were considerably heavier than other members of the group.

Table 3: Group values for weight/height indices ($\frac{Wt}{Ht^2}$) at start and finish of diet^a

	Pea group	Cornflakes group	"t" test	SE
At start	2.41	2.54	NS	± 0.06
At finish	2.42	2.54	NS	± 0.05
"t" test	NS	NS		
SE	± 0.02	± 0.01		

^aWt in g. Ht in cm.

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