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Wheat fibre, lactulose and rectal mucosal proliferation in individuals with a family history of colorectal cancer

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In a single-blind study 38 individuals at increased risk of developing colorectal cancer because of a family history of the disease were randomized to take 10·5 g wheat fibre (Trifyba) or 60 ml lactulose daily for 12 weeks. Rectal biopsies were taken before and after treatment and rectal mucosal proliferation was measured by the crypt cell production rate (CCPR). The mean(s.d.) CCPR was significantly lower in those taking wheat fibre after 12 weeks (7·2(3·4) crypt cells per crypt per h) compared both with values obtained before treatment with wheat fibre (10·2(5·1) crypt cells per crypt per h; $P = 0·02$) and after treatment with lactulose (9·4(3·8) crypt cells per crypt per h; $P = 0·05$). Proliferation in the lactulose group was not significantly different at 12 weeks compared with the value obtained before treatment. This study confirms an anti-proliferative effect of wheat fibre in a group of high-risk individuals.

It is becoming clear that relatives of individuals with colorectal cancer are at increased risk of developing the disease[1–3]. Rectal mucosal proliferation in these individuals is increased and thought to be a potential risk marker[4–7]. The proportion of individuals within a family demonstrating hyperproliferation depends on the degree of risk[4]. At present there have been few studies investigating the interaction between diet and heredity in this group. The genetic basis for the development of cancer depends on damage to the genome, which may be hereditary as in familial adenomatous polyposis or caused by environmental influences such as components of the diet. The development of cancer is dependent on the amount of genetic damage, which occurs in a stepwise fashion[8]. The mechanism of genetic damage by the faecal stream is still not clearly understood and dietary advice is limited and unclear.

Burkitt *et al.*[9] suggested that dietary fibre may protect against colorectal cancer due to a reduction in transit time and by faecal dilution of carcinogens. Epidemiological dietary studies, including a recent meta-analysis of 13 dietary studies, support a protective effect for dietary fibre in colorectal cancer[10–12]. The reasons for the apparent protective effect of fibre are unclear. Fibre may exert its effect when metabolized to short-chain fatty acids, which reduce colonic pH. Conversion of primary to secondary bile acids is regulated by the enzyme 7α-dehydroxylase found in the gut bacteria. This enzyme is inhibited below pH 6·5[13], a level commonly found in individuals at low risk of colorectal cancer, who have a faecal pH at least 0·5 units lower than those at high risk or with cancer[14,15].

Carcinogen-treated rodents with acidified colons produce significantly fewer tumours than controls[16]. In some reports acidification by the addition of fibre increased colorectal mucosal proliferation[17,18]. In contrast, other studies have suggested that high-fibre diets in rodents either have no effect on proliferation in the distal colon and only a minimal effect on the proximal colon[19] or they decrease mucosal proliferation[20].

Kashtan *et al.*[21] investigated 50 individuals who had had adenomas and were given either placebo or sodium sulphate to acidify the colon. There was a fall in pH of 0·3 but no change in rectal mucosal proliferation in the treated group. It has been shown that lactulose reduces secondary bile acid formation in the faeces of individuals with adenomas. This effect may be due to acidification of the colon or to decreased colonic transit time. However, mucosal proliferation was not measured in these studies[22–25].

In a recent dietary intervention study, Reddy and colleagues[26] found that wheat fibre produces the greatest degree of faecal dilution of bile acids when compared with oat and corn fibre. Unfortunately mucosal proliferation was not studied. However, two other studies[27,28] both showed that mucosal proliferation is decreased after the administration of wheat bran.

A study was undertaken on the effect of lactulose (alteration of acidity) and of wheat fibre (faecal dilution) on rectal mucosal proliferation in individuals at high risk of colorectal cancer because of a family history of the disease.

Patients and methods

Study group

The study group comprised 38 individuals (24 women) with a lifetime risk of colorectal cancer[1,4] greater than 1:10 and a mean(s.d.) age of 43·6(17·2) years. They were well matched for age and degree of lifetime risk. All had undergone colonoscopy and were free from neoplasia at the time of entry to the study. Patients were excluded if they were taking aspirin or other non-steroidal anti-inflammatory drugs, steroids, laxatives, calcium, vitamin D, fish oil, had colitis or were pregnant.

Study design

The patients were randomized in blocks to either 10·5 g wheat fibre (Trifyba; Sanofi Winthrop, Guildford, UK) daily or 20 ml lactulose (Lactulose; Duphar, Southampton, UK) three times daily. The investigator was not aware of the treatment. On the initial visit participants consented to both supplements and only when they reached the pharmacy were they aware which supplement they were to have. At the initial visit the participants underwent rigid sigmoidoscopy without bowel preparation, rectal pH was measured and a biopsy was taken for the measurement of rectal mucosal proliferation 8 cm from the anal verge. The biopsy containers were coded and returned to the laboratories of the Cancer Research Campaign. The biopsy was repeated at 12 weeks. Compliance was

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assessed by interview and by patients returning the material that they did not use. Good compliance was defined as usage greater than 80 per cent and poor compliance as that below 80 per cent.

Rectal mucosal proliferation

Proliferation was measured using the *in vitro* metaphase arrest technique to measure crypt cell production rate (CCPR) as previously described[4,29,30]. Briefly, explants were placed into tissue culture medium RPMI 1640 to which was added gentamicin 0·001 per cent and fetal calf serum 10 per cent. The samples were stored overnight to allow for extraction artefact[30]. At the time of assay the culture medium was replaced with fresh medium containing 1 ml 5 μg/ml vincristine. The culture medium containing the explants was incubated in an atmosphere of 5 per cent carbon dioxide and 95 per cent oxygen. The explants were removed from tissue culture after 25, 50 and 75 min, fixed in Carnoy's solution and stored in 70 per cent ethanol. The number of metaphase arrests was counted in 20–30 crypts. The CCPR was calculated by least squares regression analysis and expressed as crypt cells per crypt per h.

Rectal pH

Rectal pH was measured using an oesophageal pH probe, calibrated to read between pH 9·2 and 3·0. The pH probe (Zinetics, Oxford, UK) was an antimony graphite electrode. The earth electrode was attached to the patient's skin.

Statistical analysis

A 90 per cent efficacy was postulated at a power of 80 per cent. Power calculation showed that a sample size of 38 individuals was required to show a difference with a significance level of $P = 0·05$. Comparisons between groups were made by paired Student's t test and linear regression analysis. Data are given as mean(s.d.).

Results

One individual in the wheat fibre group (telephone interview) withdrew from the study. Six patients had taken less than 80 per cent of the supplements (wheat fibre, two; lactulose, four), substantiated by returned medication in four individuals. In the wheat fibre group, three individuals had an increase in bowel frequency and six said they had had an increase in stool volume. In the lactulose group, 11 individuals reported gastrointestinal cramps and bloating, while seven reported diarrhoea and an increase in stool frequency.

There was a small fall in pH in both groups that was not statistically significant. After lactulose the pH fell from 6·6(1·2) to 6·3(1·2) and after treatment with wheat fibre the fall was from 6·7(1·0) to 6·4(1·0). The CCPR was lower after 12 weeks of wheat fibre: 7·2(3·4) *versus* 10·2(5·1) crypt cells per crypt per h before treatment ($t = 2·47$, 34 d.f., $P = 0·02$) and 9·4(3·8) crypt cells per crypt per h after 12 weeks of lactulose ($t = 2·03$, 35 d.f., $P = 0·05$). Proliferation in the lactulose group was not significantly changed after treatment (*Fig. 1*). Thirteen of the 18 individuals taking wheat fibre had a fall in CCPR although four individuals had a positive change and one had no change in proliferation. Eleven of 19 subjects receiving lactulose had a positive change or no change in CCPR. There was no correlation between the percentage change in proliferation and the change in pH in either the wheat fibre group ($r = 0·27$) or the lactulose group ($r = 0·20$).

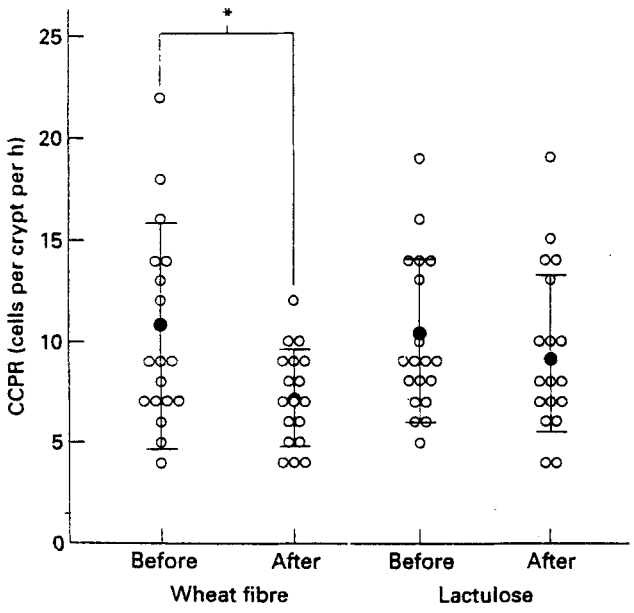

Fig. 1 Crypt cell production rate (CCPR) in subjects at increased risk of colorectal cancer before and after 12 weeks of therapy with wheat fibre or lactulose. Bars are mean(s.d.). *$t = 2·47$, 34 d.f., $P = 0·02$

Discussion

The primary endpoint of this study was a reduction in CCPR. The dose of lactulose used was expected[31] to reduce pH by 0·5 unit. The mean fall in pH in the lactulose group was 0·46. In the wheat fibre group there was a mean fall of pH 0·26. This may be due to degradation of the wheat fibre to short-chain fatty acids. Despite the fall in pH after lactulose, no effect was seen on mucosal proliferation. These findings are similar to those of Gregoire *et al.*[32], in whose study normal low-risk volunteers were given wheat bran as bread and no decrease in proliferation was found.

It has been demonstrated[22–25] that the administration of lactulose can lower the levels of secondary faecal bile acids and may therefore reduce rectal mucosal proliferation. Faecal bile acids were not measured in the present study but a similar dose of lactulose was used to that in previous studies; it is therefore possible that secondary bile acids are not important in the induction of hyperproliferation.

Other studies[27,28] have shown a significant reduction in rectal proliferation by wheat fibre, which seems to be independent of changes in rectal pH. This may be due to faecal dilution or decreased colonic transit. An alternative explanation for the reduction in mucosal proliferation is the production of short-chain fatty acids such as butyrate which are thought to be the fuel of colonic mucosa and are absorbed by both passive and active transport systems[33]. Butyrate and short-chain fatty acids may have a pH-independent antineoplastic role in colorectal cancer.

Wheat fibre meets a number of desirable criteria if it is to be used as a cancer preventive agent on a large scale. It has few side-effects as it is insoluble fibre, and has a high concentration of fibre to minimize the volume. The contents are standardized and with minimal levels of undesirable components. Lactulose is a synthetic disaccharide that passes into the colon where it is metabolized to lactic and acetic acids, and later to hydrogen and methane[34]. The osmotic effect of unabsorbed disaccharide produces gastrointestinal disturbances.

This study confirms an antiproliferative effect of wheat fibre in a high-risk group and suggests a role for supplementation in the ever-increasing number of

individuals at increased risk of colorectal cancer because of a family history.

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