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Progress report

Large bowel cancer: Causation and management

The death rate from large bowel cancer in England and Wales has tended to fall recently[1] but the change, which is slight, is probably accounted for by improvements in surgical and anaesthetic techniques. In contrast, incidence rates, if not actually rising are probably stationary. For a disease which is commonly considered curable, the five-year mortality rate, which is usually in excess of 60%, is disappointingly high. Simple technical advances in surgical management seem unlikely to contribute more than marginally to lowering death rates further since the scope for radical changes seems very limited. Hope for the future seems to depend upon progress in three fields: the identification of predisposing factors, the development of better diagnostic techniques, and the introduction of effective chemotherapy.

The incidence of large bowel cancer varies by approximately ten-fold from one part of the world to another, and broadly speaking its frequency is greater in developed countries, especially in western Europe and North America, than in South America, Africa and Asia[2,3,4,6]. However, even within the developed areas there may be considerable variations; thus Scotland has a particularly high incidence of rectal cancer, whilst in Scandinavia large bowel cancer is considerably more common in Denmark than in Finland[5]. There is also a general inverse correlation, at least within developed countries, between large bowel and stomach cancer frequency.[1] These patterns seem to have altered very little recently although the expectation of the disease in migrants from one area to another changes within one generation from that found in the old country to that of the adopted area. Thus there are marked increases in large bowel cancer frequency in Japanese who move to the USA and in Puerto Ricans who do likewise[6]. Gastric cancer incidence rates seem to fall coincidentally but rather more slowly. The environmental influences responsible for these changes are still obscure. Comparisons of diets and social characteristics of colonic cancer patients and matched controls have, in general, been disappointing in their outcome[7,8], and therefore comparisons of broad dietary and other characteristics in areas with high and low incidence rates have been attempted instead. Of the many factors studied, only dietary constituents, particularly the proportions of animal protein and fat, have been shown to correlate well with the geographical variations in the incidence of large bowel cancer[6,7,8,9,10,23]. Thus, 12% of calories in the diet in Japan, a low-risk area, seem to be derived from fat (mostly unsaturated) whilst in the USA almost 40% has been calculated as being derived from fat, of which almost half is saturated[7]. The Japanese in the USA seem to adopt a western diet whilst in Japan itself patients with colonic cancer seem to adhere to a western diet more often than controls[6]. The strong positive correlation between international mortality rates from colonic cancer and coronary heart disease, a disease in which a connexion with fat intake is more generally accepted, supports the hypothesis linking dietary fat with the aetiology

of colonic cancer, as do some case control studies. However, it is also worth noting that within western countries no correlation has been found between serum cholesterol concentration and the later occurrence of colonic cancer[11].

Available data on beef consumption support an alternative hypothesis linking the incidence of colonic cancer with animal protein intake. Argentina, New Zealand and Uruguay are three countries with particularly high consumption rates for beef. New Zealand has an extremely high national incidence rate for bowel cancer[5] whilst the mortality rate in Argentina, a relatively impoverished country, is as high as that of the USA[1]. Finally, the incidence rate projected from mortality data for Uruguay also seem to be very high[4]. There may also be significance in evidence that in Scotland, where the incidence is high, it is particularly so in rural areas and these include the beef-raising counties such as Aberdeenshire[13].

An attractive general explanation of such observations is that dietary constituents influence the microbial flora of the gut and/or intestinal secretory patterns. The structural similarity of bile acids to carcinogenic polycyclic hydrocarbons[14], the synthetic pathway which can be devised from deoxycholic acid to a carcinogen, such as 3 methyl cholanthrene[15], and the carcinogenic potential in animals of deoxycholic acid, bisnor 5 cholenic acid and apocholic acid[16,17,18], give specific supporting evidence to such a theory even though these steroidal molecules do not seem to cause adenocarcinoma in the bowel experimentally. Bile acids are metabolized by anaerobic organisms such as Bacteroides and Clostridia species. Comparisons of the faecal bacterial flora of English and Ugandan adults showed that more bacteroides and fewer streptococci and lactobacilli were detectable in specimens obtained from the English[19], and similar trends were found in a wider comparison in three countries with a low incidence of colonic cancer (Uganda, India and Japan) and three with high incidences (England, Scotland and the USA)[15]. In addition, a strong correlation was detectable between faecal concentrations of neutral sterols (derived from cholesterol), acid sterols (derived from bile acids) and the incidence of colonic cancer[15].

Broadly, confirmatory evidence has been obtained in the USA. Faecal samples obtained from individuals on ordinary diets were found to contain greater amounts of coprostanol and coprostanone (products of cholesterol degradation), lithocholic, deoxycholic and total bile acids, and β glucuronidase (as a measure of microbial activity) than did samples obtained from Seventh Day Adventists not taking animal protein and from Japanese and Chinese in the USA taking traditional diets[20]. Such studies demonstrate that the microbial flora of the gut does vary in association with dietary changes and that the faecal excretion of substances of biliary origin also varies in parallel.

Proof that such variations are significant in inducing cancer in man has yet to be obtained. Higher bile acid concentrations and large numbers of clostridial organisms have been found in the faeces of patients with large bowel cancer compared with those with other diseases, but since bowel cancer commonly alters bowel habit, cause and effect are impossible to distinguish[21]. Large-scale prospective studies could supply an answer but if the expectation of colonic cancer in the middle-aged and elderly lies between 50 and 100 per 100 000 per year, then, even leaving aside a run in period of say five years to exclude present but undetected bowel cancer, the number of samples required is daunting.

A reduced dietary fibre intake has also been suggested as an important factor in the incidence of large bowel cancer[22]. Bowel frequency in rural Africans has long been known to be greater in such individuals than in western populations, and intestinal obstruction due to volvulus (perhaps due to the increase in colonic size in persons on a high-fibre diet) is common, whereas intestinal cancer seems to be rare. Such characteristics have been emphasized by Burkitt[3], and it has been propounded that a high dietary fibre content might be beneficial because it might reduce intestinal transit time and hence the length of time any carcinogen can remain in contact with the bowel wall. Secondly, it has been suggested that a high dietary fibre intake might reduce bile salt degradation[23]. Support for the fibre hypothesis is hard to produce because the characteristics of fibre itself are difficult to identify and quantify. Dietary fibre is heterogeneous and the simple definition of crude fibre as that portion which is left after acid and alkali reduction, is clearly inadequate. However, at least one piece of evidence suggests that a simple inverse correlation between transit rate and bowel cancer frequency is untenable. The Japanese in Hawaii have been found to have a lower cancer incidence than the Americans there, but the Japanese bowel transit time is, if anything, slower[24].

Most large bowel cancers have been considered in the past to be within reach of the examining finger or sigmoidoscope but recent North American evidence suggests that right-sided cancer may be becoming increasingly frequent[25]. If this is generally true, then conventional diagnostic methods will have to rely heavily upon contrast radiography and upon colonoscopy.

The failure of established diagnostic methods to affect the mortality rates in this disease materially has nurtured the hope that something useful will emerge from research in the field of tumour immunology.

Carcinoembryonic antigen (CEA), first identified in 1965 in the serum of patients with large bowel cancer[26], has been disappointing in its application as a diagnostic test. Although almost all patients with disseminated disease have raised values, they are raised in less than 60% when the disease is localized[27,28,29]. Furthermore, it is not tumour-specific as was originally thought, and is found in variable amounts in a variety of other neoplastic[29,30] and non-neoplastic[31,32] disorders. In addition, its estimation in ulcerative colitis does not seem to discriminate between those with and without early malignant change[33]. Its major application, if any, is in postoperative follow up since it allows the detection of recurrent tumour several months before it becomes clinically evident[34,35,36].

Cell-mediated immune reactions have also been under extensive investigation, the rationale being first that tumour cells express antigenic determinants, which are not found in normal cells of the same tissue and are thus immunogenic[37]; and secondly, in that an inverse relationship exists between the degree of lymphocytic infiltration in tumour tissue, and the presence of cellular undifferentation or metastatic spread, thus suggesting a role for sensitized lymphocytes in limiting tumour spread[38].

The ability of cancer patients to develop an immune response to tumours has been studied by a number of techniques *in vivo* and *in vitro*. The results have varied according to the methods used both in preparing the tumour extracts and in demonstrating specific responses.

The approach *in vivo* has been to skin test with soluble tumour extracts, observing the delayed cutaneous response. In one study, less than half the

patients with colonic cancer had a positive response[39], whilst others have found a much higher rate of positivity[40]. More recent work has shown that patients with colonic cancer generally give a positive skin test to extracts of fetal large bowel but not to similar extracts from adult colon[41], whereas such reactions are undetectable in control patients with other primary tumours. The antigens involved in these reactions seem to be distinct from CEA.

The ability of sensitized mononuclear cells from patients with cancer to inhibit the growth of, or kill, tumour target cells maintained *in vitro* has formed the basis of the colony inhibition test[42] and microcytotoxicity assay[43, 44] respectively. These have been applied extensively to the study of large bowel cancer, and tumour-directed immune reactivity has been demonstrated in a high proportion of patients. It has been shown that cytotoxic cells are active against both autochthonous and allogeneic target cells provided they come from tumours of a similar histological origin, implying that such tumours share type-specific neo-antigens[45,46,47]. This cytotoxicity can be abrogated by the addition of serum from tumour-bearing patients[48] or of extracts prepared from a homogenate of the tumour tissue[49]. The nature of this 'blocking factor' is uncertain but its presence in the serum of patients with cancer may in part explain the ineffectiveness of their circulating, tumour-directed cytotoxic lymphocytes. Doubt has been shed on the specificity of these reactions in extensive studies by Takasugi *et al*[50,51], but the complexity of these methods make it very unlikely that they could ever be adapted to be of use in a diagnostic setting. Two other simpler methods have been used to detect sensitization of lymphocytes to tumour-specific antigens. They rely on the observation that sensitized lymphocytes release a number of soluble factors (or lymphokines) on contact with the appropriate antigen. One, the migration inhibition factor is detected by its effect on the movement of peripheral blood leucocytes in an *in-vitro* assay system. The migration of leucocytes taken from patients with large bowel cancer is retarded in the presence of the supernatant from a crudely prepared homogenate of allogeneic colon cancer tissue, an effect not seen in patients without colonic or rectal cancer[52,53,54]. A blocking effect of tumour-bearer serum has again been described[53]. This migration inhibition does not occur to the same extent in response to more refined tumour extracts (for example, 3M KCl or papain-treated material which contains solubilized and soluble constituents of the tumour cell or cell membrane respectively)[55]; or to crude extracts prepared from normal colonic mucosa[53,54]. Though leucocyte migration inhibition is often detectable in samples obtainable from patients with limited tumour spread, making it potentially more promising than CEA in early cancer detection, the effect is often small and sometimes undectable at all. It thus has great limitations as an additional clinical investigation system unless the cause of non-responsiveness can be identified, and the degree of inhibition enhanced. A second *in-vitro* test under investigation is leucocyte adherence inhibition[56]. It depends basically upon the observation that normal adherence to glass of peripheral blood leucocytes is inhibited if the leucocyte preparation is drawn from a cancer patient and is exposed first to a tumour extract of similar type. Preliminary studies have shown specific inhibition of adherence after exposing peripheral blood leucocytes from patients with colonic cancer to colonic cancer extracts but this has yet to be confirmed on a wider scale[57].

Whilst these experimental studies are of considerable interest, they are neither sufficiently reliable nor discriminative to be of value diagnostically in

ordinary clinical practice.

Prospects for improving treatment by non-surgical means remain far distant. Available chemotherapeutic agents are largely ineffective and there is, as yet, no evidence that adjuvant immunotherapy will be clinically useful; this whole field is ill developed and much more work is needed.

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