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Epidemiologic Studies of Diet and Cancer[1] (42420)

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The hypothesis I want to discuss have to do with fats, fiber, and vitamin A. First, a word about the various methods of study, particularly in epidemiology. Experiment is the scientific instrument par excellence and as utilized in intervention trials in epidemiology has been called the most powerful epidemiologic tool. An example in dietary epidemiology is the current trial going on among American doctors whereby they are taking carotene or a placebo on a random basis.

The criteria for intervention trials have to be observed very carefully: there has to be evidence in favor of a salutary effect of the substance being tried. There has to be evidence that there is no long-term toxicity, and there has to be the likelihood that the human participants will comply with the stated regimen for years, sometimes, and that the individuals on the placebo will stay on the placebo. For various reasons, past large-scale human intervention trials, although costing many millions of dollars and absorbing huge amounts of time of investigators, have yielded convincing results in only a few instances.

The least powerful tool of epidemiology is the international correlational study. I don't have to dwell on the drawbacks of these. A typical study relates the mortality from colon cancer to the ingestion of fats in populations in different countries (1). Even though there is a correlation, this does not prove that fats cause colon cancer. The proportion of people involved in manufacturing can be related to colon cancer in a similar fashion. There is growing evidence that exercise reduces risk of colon cancer. We might hypothesize that those nations requiring a great deal of physical labor; that is, those nations in which large proportions are not involved in manufacturing subject their populations to much more exercise, and this could account for their low mortality from colon cancer.

A number of people have suggested the study of nutrients in biological materials to examine their epidemiological role. This has the potential for contributing a good deal to the total of epidemiological evidence, just as the other two methods do. But even here there are difficulties. Among other things, we are interested in the level of serum nutrients because they may reflect the human behavior responsible for them. If we are able to identify dietary behavior which might raise or lower the level of a given nutrient in the serum, and which is also related to risk, we may have evidence that this behavior should be changed, that is, either eliminated or intensified. The difficulty is, however, that serum nutrients do not necessarily reflect dietary behavior. Walter Willett's inquiry comparing plasma carotene with that reported in a questionnarie, indicated a fair amount of correlation, around 0.30 (2). The same was true of α-tocopherol, but no correlation was found between dietary retinol and serum retinol. Hyman did a similar study and found little correlation between serum cholesterol and cholesterol as reported in survey research interviews or 3-day food records (3). When one considers that the level of serum nutrients further might be altered by the disease or by the therapy for the disease in case-control studies, one can see other problems. There are ways of making this tool productive, however, and we may get to these later.

One way out of the difficulty is a prospective design; that is, you take blood samples, measure the level of cholesterol or other nutrient, and then examine the incidence of disease in individuals with various levels of the nutrient—in this case, cholesterol. An example is the famous cohort, that from Framingham, Massachusetts (4). Of course, these studies cost tremendous amounts of money and take enormous amounts of time. In some senses they are definitive, but they rarely yield enough numbers for multivariate analyses.

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Another approach to study is the analysis of reported diets by interviews and questionnaires in either case-control studies, retrospectively, or in prospective studies. There are methods of interviewing that can get at dietary behavior in an approximate but useful way. There is a good deal of evidence for this. In 1963, we did our first reinterview study where we compared the answers of individuals to a food frequency questionnaire to what they had said on the same questionnaire an average of 18 months earlier (5). On the average, about 89% of the answers on the second interview reflected those on the first. More recently, we compared answers of men with those of their wives as to the men's dietary behavior, and found that the proportion of spouses agreeing within one category ranged from 77 to 95% (6). There was not a great deal of difference between case–spouse pairs and control–spouse pairs.

These results are similar to those found by Kolonel and his colleagues in Hawaii (7). There have been a fair number of other inquiries into the reliability of diet studies and they concur in assessing the method adequate to assess dose–response relationships (8, 9).

A very serious problem with dietary questionnaires is that the most cogent diet for the test of hypotheses may be that throughout life. It would include the diet at the time a disease might have been initiated, maybe two decades ago, as well as the diet a year or two prior to the onset of symptoms. Thus, the investigator needs to study diet at various times in life. In 1982, Tim Byers reinterviewed individuals we had first interviewed in the period 17 to 25 years earlier (10). He found a high correlation between the original diet and the current diet, but that a better correlation with original diet is the current recollection of the original diet. Thus, in interview studies it is important to assess changes in diet throughout life.

Another approach hallowed by tradition is to use animals by studying the way they respond to exposure to given substances. The difficulties of generalizing from species to species are well known. A couple of striking examples in dietary epidemiology are in the studies of saccharine, which readily produces tumors in animals, but which has yet to produce tumors in people. When Robert Hoover did a large, nationwide study of the relationship between bladder cancer and uses of primarily saccharine, he found no relationship (11). Another instance of a contradiction between human and animal findings is in the study of alcohol and cancer of the mouth, esophagus, and larynx. We found an increase in risk of oral cancer associated with an increase in alcohol consumption (12). A similar finding was obtained in studies in cultures as disparate as a group in Puerto Rico (13), in Paris (14), a couple of populations in New York City (15), and in Brittany (16). Nonetheless, no one has been able to produce tumors in animals with alcohol. Not even Fred Bock, who tried it with bourbon (17). I would image that most epidemiologists and nonepidemiologists, however, would buy the finding in humans in spite of the negative finding in animals.

So all of the various approaches to the study of human nutrition and risk of cancer have some drawbacks. On the other hand, they all have some advantages, and I think it is important in resolving this dilemma to look for corroboration from studies of a variety of sorts. Where this has not been possible, as in the relationship of alcohol and cancer of the upper alimentary tract, I think we must rely on extensive replication in humans to nail the finding down.

The first of the hypotheses I want to discuss has to do with fiber and colon cancer. Dennis Burkitt first promulgated the notion that a high-fiber diet speeds transit time, which lessens the amount of time any carcinogen in the fecal stream might have contact with the gut lumen. Moreover, high fiber increases bulk and dilutes any carcinogen present. He based this hypothesis on the observation that Africans have much less colon cancer than the English, and that African school boys have a lot more fiber in their diets than English school boys (18). The fact that he was generalizing from a very young age group to a very old age group, from one race to another, and from one culture to another did not deter him. But there is a certain logic to the hypothesis. Fiber does seem to dilute the fecal stream, but transit time seems to be affected in various ways by fiber. We will hear more later regarding the production of tumors in animals with fiber.

The most convincing epidemiologic study is by Baruch Modan, in Isreal (19). I helped.

We found that colon cancer cases ate significantly less fibrous foods than either neighborhood controls or surgical controls in five hospitals in Tel Aviv. However, there are relatively few other convincing studies. Bjelke seemed to find a small relationship (20), and Dales in a study of 77 Blacks in the Bay Area found that persons with low fiber and high fats ingestion had a two-fold risk (21). Our own study, recently concluded, compared cases from Erie, Niagara, and Monroe counties, the three counties of Buffalo, Niagara Falls, and Rochester. We found no relationship between the dietary fiber ingested per month and the risk of cancer of the colon in either males or females (22). Our logistic analyses examining fiber adjusted for fats and calories found, however, a significantly decreased risk. This is not unlike Dales' finding. A close examination of risk associated with fiber in deciles of consumption of fats suggested that fiber seems to have a protective effect in individuals with low and moderate exposures to fat but has no effect given high exposure to fat. Despite the real paucity of case control or prospective data linking dietary fiber to colon cancer, the hypothesis persists in even what might be called the best circles, perhaps because of its simple logic as well as the amount of publicity it has received. Of course, other nutrients are found in the same foods that furnish fiber, for example, vitamin A.

Vitamin A might be hypothesized to reduce risk of cancer because of its effect on squamous epithelial tissue. Lack of vitamin A compromises the integrity of such tissue, perhaps through increasing the permeability of cellular walls and their accessability to a carcinogen. Studies with animals will be examined later.

One of the first studies in epidemiology done on this subject was a cohort study of some 8000 Norwegian males in which their diets were characterized by Eric Bjelke, using a vitamin A index (20). Dr. Bjelke compared those on a low and high index of vitamin A who smoked various amounts of cigarettes, and he found that the incidence of lung cancer in these individuals varied very substantially in terms of the amount of vitamin A they reported in their diets. For those who had ever smoked, the incidence was about 7 per 1000 in those ingesting a low amount of vitamin A as compared to 1.7 per 1000 in those ingesting a large amount. Similar differences with the two levels of vitamin A occurred throughout. Our case-control study among Roswell Park patients came up with a similar finding (23). The smoking and age-adjusted relative risk of cancer of the lung decreased with increases in level of dietary vitamin A ingested. High levels of retinoids, mainly carotene, in the diet were involved in statistically significant decreases in risk for cancer of the lung, bladder, mouth, larynx, breast, cervix, and ovary. There was no effect for the gastrointestinal cancers or cancer of the endometrium. Note especially that there is an increase in risk of cancer of the prostate.

The dietary questionnaire which we used in those days was primitive and contained questions dealing mainly with vegetable sources of vitamin A. Note that many of the cancers for which the carotene seemed to decrease risk were cancers of squamous cell types. Tim Byers, as a result, was moved to examine the relationship between dietary vitamin A and lung cancer of squamous cell and adenocarcinoma types (24). He found the expected inhibition of risk with increases in ingestion of vitamin A for squamous cell carcinoma of the lung but not for adenocarcinoma of the lung. Our more recent studies in the three western New York counties indicated again that carotene, but not preformed vitamin A, decreased risk for cancer of the lung, larynx, and bladder (25).

The anomaly in all of this was revealed when we examined the relative risk of prostate cancer by monthly ingestion of a number of nutrients and particularly vitamin A. We found that as ingestion of vitamin A increases from the lowest to the highest frequency, the risk of cancer of the prostate increases significantly. Larry Kolonel at the University of Hawaii found the same thing. Heshmat *et al.* had a similar finding in a study of 188 Blacks in Washington, D.C. (27). Although the contradiction is disturbing, it may not be a contradiction at all. I see no reason why a given anticarcinogen necessarily must be an anticarcinogen for all types of cancer. Indeed, various retinoids have been found to enhance tumors in animals under certain special circumstances. The potential vis-à-vis increasing risk in humans is especially disturbing. We obviously need to study this further, and as soon

as possible, to determine whether there might be a true enhancement of risk of cancer of the prostate and the other diseases with increases in ingestion of carotene.

The final hypothesis which I wish to discuss with you is that dietary fats might increase risk of cancer at a number of sites.

A number of epidemiologic serum cholesterol studies have been done in prospective settings. For example, Robert Hiatt studied the subsequent incidence of cancer of the breast in 87,000 women who had their serum cholesterol measured earlier and found no difference in incidents with increases in serum cholesterol (28). Similarly, Manning Feinleib summarized results of a large number of prospective studies which assessed serum cholesterol at the beginning of the study and measured the subsequent cancer mortality among those individuals exhibiting various levels of serum cholesterol (29). He found an inverse relationship in these particular studies, that is, an increase in serum cholesterol was associated with a decrease in total cancer mortality. In Framingham, this was true even after subtracting cases which developed within 10 years of taking the bloods (4). In an equally large number of studies, he found no relationship (29). In no instance did he find that an increase in serum cholesterol was related to an increase in incidence of cancer.

One of the earliest dietary studies was that done by Haenszel among the Japanese of Hawaii and the Japanse of Japan (30). He obtained contradictory results. Thus, the Hawaiian Japanese, who frequently ingested beef had a twofold relative risk as compared to those who infrequently ingested beef. In Japan, however, no such difference was observed. The same thing was found for total meat and pork. On the other hand, there are a large number of studies which are positive. Lubin *et al.* found an increase in risk of breast cancer with increases in ingestion of meats and fish, beef and pork, and total animal fat (31). All of this was indicated in a very brief dietary questionnaire. They found no difference for foods containing cholesterol. Hinds *et al.* did a similar study of cancer of the lung as related to dietary cholesterol and found an increase in risk with an increase in ingestion as measured by an index based upon reported diet (32).

Results of our study of cancer of the cervix showed increased risk associated with increased ingestion of animal fat (33). Also, individuals eating high levels of carotene were at only 22% of the risk of individuals not doing so. In our study of cancer of the prostate in RPMI patients, there was an increase in risk with increases in ingestion of animal fats for both the younger and the older patients (34).

In the case of cancer of the esophagus, two studies, one by Ziegler on 120 Blacks in Washington, D.C. (35) and another by Tuyns on 740 cases and 1300 controls in Calvados, France (36), showed decreases in risk with increasing ingestion of fresh meat and fish and vegetables and fruits, adjusted for tobacco and alcohol ingestion. This replication of findings at oods with the current fat hypothesis in two very careful and sophisticated studies suggests either there is an exception to the theory for this specific site or that the fats hypothesis in general needs further examination.

Our current study, which is now in the process of analysis, based upon the three western New York counties, shows an increase in risk of cancer of the mouth and colon with an increase in fats ingestion. The relationship is not simple, however. To focus on cancer of the colon specifically, fats, calories, obesity, and caloric expenditure may interact in a complex fashion to affect risk. Risk of cancer of the colon increases with an increase in the ingestion of fats per month as reported on dietary interviews. Moreover, there was an increase in risk of cancer of the colon with increases in ingestion of total calories.

One might hypothesize on the basis of these data that the obese individual would have the higher risk of cancer. Indeed, as the Quetelet Index increased, suggesting higher obesity, the risk of colon cancer similarly increased both for males and females. When we examined the risk associated with obesity adjusted for fats, calories, and fiber in a logistic analysis, obesity continued to be related to higher risk of cancer of the colon. Thus, it could be that the ingestion of calories not expended may lead to the obesity which carries higher risk. This suggests the potential importance of physical exericse.

There have now been five inquiries into the relationship between exercise and risk of colon

cancer, including our two (37–41). Our first study examined the relationship between colon cancer and exercise required on all jobs held throughout life. As the proportion of life in sedentary activity increased, so did the risk of cancer of the colon, adjusted for age (37). In our second inquiry, we found something similar in the Washington State population (38). In females we found a reduction in risk of breast and colon cancer associated with increased physical activity. Exercise thus may function to reduce risk, but this certainly needs further replication.

In a very impressionistic way, my own assessment of the current state of knowledge with regard to fats and carotene is as follows: It would appear that there are a very few studies suggesting that fats increase risk of cancer of the mouth, larynx, lung, colon, bladder, prostate, cervix, and breast, and there are a very few studies suggesting that carotene is associated with decreases in risk of cancer of the mouth, larynx, esophagus, lung, bladder, cervix, ovary, and breast. However, in almost every case, the risks associated with these factors are small, ranging from 1.8 to 2.5.

There is very little replication of the relationship between diet and a number of very important sites such as breast, prostate, colon, and lung cancer, and none on less frequently occurring cancers such as melanoma and cancer of the mouth, cervix, bladder, endometrium, and ovary. Again, there are very important anomalies, particularly the possibility that carotene may increase risk of prostate cancer and the decrease in risk of esophageal cancer with increases in fats ingestion. We might ask ourselves whether some apparent nutrient villians might decrease risk in some situations. One could ask why has gastric cancer decreased over the last three of four decades in such precipitous fashion.

The finding with regard to the Quetelet Index, suggesting an increase in risk of colon cancer with an increase in obesity, has been found in the past also for cancer of the breast and endometrium. It needs to be investigated for a large number of different cancers, and this implies that we need to conduct further studies of the relationship between exercise, calories ingestion, fats ingestion, obesity, and disease. Exercise, more and more, is beginning to appear as a panacea, just as fats may be a culprit, in the risk for coronary heart disease as well as cancer. As so often in modern epidemiology, we are again involved in multifactoral etiologies. We have more and more to be aware of the interaction of factors to either enhance or inhibit risk.

1. Wynder E. The epidemiology of large bowel cancer. Cancer Res **35**:3388–3394, 1975.
2. Willett W, Stampfer M, Underwood B, *et al.* Validation of a dietary questionnaire with plasma carotenoid and alpha-tocopherol levels. Amer J Clin Nutr **38**:631–639, 1983.
3. Hyman H, Insull W, Palmer R, *et al.* Assessing methods for measuring compliance with a fat-controlled diet. Amer J Pub Health **72**:152–160, 1982.
4. Sorlie P, Feinleib M. The serum cholesterol–cancer relationship: An analysis of time trends in the Framingham Study. J Natl Cancer Inst **69**:989–996, 1982.
5. Graham S, Lilienfeld A, Tidings J. Dietary and purgation factors in the epidemiology of gastric cancer. Cancer **20**:2224–2234, 1967.
6. Marshall J, Priore R, Haughy B, Rzepka T, Graham S. Spouse–subject interviews and the reliability of diet studies. Amer J Epidemiol **112**:675–683, 1980.
7. Lee J, Kolonel L. Nutrient intakes of husbands and wives. Amer J Epidermol **115**:515–525, 1982.
8. Byers T, Marshall J, Fiedler R, *et al.* Assessing nutrient intake with an abbreviated dietary interview. Amer J Epidermiol **122**:41–50, 1985.
9. Willett W, Sampson L, Stampfer M, *et al.* Reproducibility and validity of a semiquantitative food frequency questionnaire. Amer J Epidemiol **122**:51–65, 1985.
10. Byers T, Rosenthal R, Marshall J, Rzepka T, Cummings M, Graham S. Dietary history from the distant past: A methodological study. Nutr Cancer **5**:69–77, 1983.
11. Hoover R, Strasser P. Artificial sweetners and human bladder cancer. Lancet, April 19:837–840, 1980.
12. Graham S, Dayal H, Rohrer T, *et al.* Dentition, diet, tobacco, and alcohol in the epidemiology of oral cancer. J Natl Cancer Inst **59**:1611–1618, 1977.
13. Martinez I. Factors associated with cancer of the esophagus, mouth and pharynx in Puerto Rico. J Natl Cancer Inst **42**:1069–1094, 1969.
14. Schwartz D, Lellouch J, Flamant R, *et al.* Alcohol et cancer: Resultats d'une enquete retrospective. Rev Franc Études Clin Biol **7**:590–604, 1962.
15. Wynder E, Bross I. Aetiological factors in mouth cancer: An approach to its prevention. Brit Med J **1**:1137–1143, 1957.
16. Tuyns A. Oesophageal cancer in non-smoking drinkers and in non-drinking smokers. Int J Cancer **32**:443–444, 1983.

17. Bock F. Personal communication, 1984.
18. Burkitt D, Walker A, Painter N. Effect of dietary fiber on stools and transit times and its role in the causation of disease. Lancet, December 30:1408–1412, 1972.
19. Modan B, Lubin F, Barell V, *et al.* The role of starches in the etiology of gastric cancer. Cancer **34**:2087–2092, 1974.
20. Bjelke E. Epidemiologic studies of cancer of the stomach, colon, and rectum, with special emphasis on the role of diet. Thesis, University of Minnesota. University Microfilms, Ann Arbor, 1973.
21. Dales L, Friedman G, Ury H, *et al.* A case-control study of relationships of diet and other traits to colorectal cancer in Blacks. Amer J Epidemiol **109**:132–144, 1978.
22. Graham S, Marshall J, Haughy B, *et al.* Epidemiology of cancer of the colon in western New York. Unpublished manuscript, 1986.
23. Mettlin C, Graham S, Swanson M. Vitamin A and lung cancer. J Natl Cancer Inst **62**:1435–1438, 1979.
24. Byers T, Vena J, Mettlin C, *et al.* Dietary vitamin A and lung cancer risk: An analysis by histologic subtypes. Amer J Epidemiol **120**:769–776, 1984.
25. Graham S. Epidemiology of retinoids and cancer. J Natl Cancer Inst **73**:1423–1428, 1984.
26. Kolonel L, personal communication, 1986.
27. Heshmat M, Kaul L, Kovi J, *et al.* Nutrition and prostate cancer: A case-control study. The Prostate **6**: 7–17, 1985.
28. Hiatt R, Friedman G, Bawol R, *et al.* Breast cancer and serum cholesterol. J Natl Cancer Inst **68**:885–889, 1982.
29. Feinleib M. Summary of a workshop on cholesterol and noncardiovascular disease mortality. Prev Med **11**:360–367, 1982.
30. Haenszel W, Berg J, Segi M, *et al.* Large-bowel cancer in Hawaiian Japanese. J Natl Cancer Inst **51**:1765–1779, 1973.
31. Lubin J, Burns P, Blot W, *et al.* Dietary factors and breast cancer risk. Int J Cancer **28**:685–689, 1981.
32. Hinds M, Kolonel L, Lee J, *et al.* Dietary cholesterol and lung cancer risk among men in Hawaii. Amer J Clin Nutr **37**:192–193, 1983.
33. Marshall J, Graham S, Byers T, *et al.* Diet and smoking in the epidemiology of cancer of the cervix. J Natl Cancer Inst **70**:847–851, 1983.
34. Graham S, Haughey B, Marshall J, *et al.* Diet in the epidemiology of carcinoma of the prostate gland. J Natl Cancer Inst **70**:687–692, 1983.
35. Ziegler R, Morris L, Blot W, *et al.* Esophageal cancer among black men in Washington, D.C. II. Role of nutrition. J Natl Cancer Inst **67**:1199–1286, 1981.
36. Tuyns A, personal communication, 1986.
37. Vena J, Graham S, Zielezny M, *et al.* Lifetime occupational exercise and colon cancer. Amer J Epidemiol **122**:357–365, 1985.
38. Vena J, Graham S, Zielezny M, *et al.* Occupational exercise and risk of cancer. Amer J Clin Nutr, in press.
39. Frisch R, Wyshak G, Albright N, *et al.* Lower prevalence of breast cancer and cancers of the reproductive system among former college athletes compared to non-athletes. Brit J Cancer **52**:885–891, 1985.
40. Garabrant D, Peters J, Mack T, *et al.* Job activity and colon cancer risk. Amer J Epidemiol **119**:1005–1014, 1984.
41. Gerhardsson M, Norell S, Kiviranta H, *et al.* Sedentary jobs and colon cancer. Amer J Epidemiol **123**:775–780, 1986.
