

# Symposium: Diet, Anthropometry and Breast Cancer: Integration of Experimental and Epidemiologic Approaches

## Perspectives on Integrating Experimental and Epidemiologic Research on Diet, Anthropometry and Breast Cancer<sup>1</sup>

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**ABSTRACT** Three perspectives on the integration of experimental and epidemiologic research on diet, anthropometry and breast cancer are presented. 1) Although body weight and height have been linked to breast cancer risk by epidemiologic research, their roles have not been directly explored with animal models. However, basic, clinical and epidemiologic research on obesity and associated metabolic alterations may be pertinent. Individual differences in the timing and magnitude of weight gain and loss during adult life need to be considered in epidemiologic studies of adiposity and breast cancer, along with individual differences in the pattern of body fat deposition, the hormonal and metabolic changes that accompany the adiposity, and family history of obesity-related chronic diseases. Animal models with genetic predispositions to obesity, diabetes and breast cancer merit further exploration, as well as models that can evaluate exposures occurring after puberty. 2) The synergy between experimental and epidemiologic studies on fat and energy intake and breast carcinogenesis has been productive because each discipline has had to incorporate recent findings of the other. Dietary studies utilizing animals with different genetic profiles are promising, but require identification of the critical genes in human carcinogenesis. 3) Controlled dietary intervention studies with human participants using intermediate endpoints can bridge the gap between animal and epidemiologic studies, but generally accepted intermediate endpoints for breast cancer need to be developed. Such studies would permit better control of diet than large clinical trials and the opportunity to explore mechanisms. *J. Nutr.* 127: 936S–939S, 1997.

**KEY WORDS:** • *animal disease models* • *biomarkers* • *breast cancer* • *dietary fat* • *weight*

### ***Dr. Ballard-Barbash: Integration of basic, clinical and epidemiologic research on anthropometry and breast cancer***

The positive association of body weight and height with postmenopausal breast cancer has been found in many studies since the early 1970s (Ballard-Barbash 1994). Early etiologic hypotheses focused on the probable role of excess nutrient or energy intake in increasing risk and cited supporting evidence from basic research in animal models that found increases in mammary tumors with feeding diets high in energy and fat. Animal models designed to explore the role of energy and fat in mammary carcinogenesis have not been designed to directly examine how weight or height might increase breast cancer risk. However, basic research using animal models of obesity,

as well as clinical and epidemiologic research on obesity and associated metabolic alterations, may stimulate the formation of new hypotheses regarding the underlying metabolic factors influencing breast cancer cell growth. These hypotheses may help to integrate data related to timing of weight gain, associated metabolic changes, and interaction with familial or genetic predisposition to disease.

Research in obesity has clearly demonstrated that it is a heterogeneous condition in terms of etiology and its association with health outcomes. The heterogeneity of its association with health outcomes is relevant to the design of epidemiologic research examining how weight and weight-related factors may influence breast cancer risk. For example, although excess weight or body mass is positively associated with risk of chronic disease, such as coronary artery disease or hypertension, among middle-aged women, these associations are less consistent among older women. That is, antecedent weight history modifies the association between body weight and coronary heart disease among older women and helps to clarify interpretation of inconsistent associations between body weight and coronary heart disease in this population (Harris et al. 1993).

Thus, weight or body mass alone may not be a good measure of risk or even of past underlying metabolic factors that influ-

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enced risk of breast cancer, particularly in studies involving older women. In the last 5–10 y a number of epidemiologic studies have shown increases in breast cancer incidence with both increased central body fat and weight gain even in cohorts that show minimal association of body mass with breast cancer incidence. Furthermore, a recent analysis by Ziegler and colleagues (1996) in a group of Asian-American women suggested that both increased body mass at the time of diagnosis and adult weight gain, particularly in the decade prior to the diagnosis of breast cancer, consistently predict increased risk for breast cancer among women aged 50–55 y. Similar to results of two other studies (Ballard-Barbash et al. 1990, Brinton and Swanson 1992), weight loss was associated with a reduction in risk that was not statistically significant. The suggestive finding that weight loss may prevent the development of breast cancer requires replication in studies specifically designed to address issues such as how timing, amount and method of weight loss influence breast cancer risk.

Individuals with different types of obesity or genetic predispositions to develop obesity demonstrate a range of metabolic responses to conditions that may increase either overall or central body fat accumulation. Differences in metabolic response have been found in both the concentration and metabolism of lipids, insulin and sex steroids. Furthermore, an increasing body of research has demonstrated that other hormonal factors, such as insulin, insulin-like growth factors and androgens in addition to estrogen, may stimulate breast cancer cell growth. When juxtaposed, these findings suggest that the underlying metabolic factors potentially explaining the observed associations of body mass, weight change and central body fat with breast cancer extend beyond excess nutrient intake or postmenopausal persistence of estrogen and should be explored in future studies. Furthermore, with the evidence that these metabolic variables are altered by multiple factors, such as changes in weight, physical activity and diet and during pregnancy and menopause, it has become evident that a more careful assessment of these multiple factors is needed to provide a more complete understanding of how these complex interrelationships, including body weight and height, influence breast cancer risk.

Finally, recent research suggests that the association of body weight and central body fat with breast cancer may be modified by a familial predisposition to hormone-dependent cancer; compared with women without a family history of breast cancer, women with a family history of breast cancer who are heavier or have more central body fat experience greater increases in risk (Sellers et al. 1992). Furthermore, among women with a family history of breast cancer, both the presence of diabetes mellitus and more central body fat increase breast cancer risk (Sellers et al. 1994). If hyperinsulinemia is one mechanism by which these conditions increase risk, it is likely that women with a familial predisposition to develop diabetes mellitus may also be at increased risk for breast cancer. To date, basic research in animal models has not been designed to address the influence of central body fat or weight change at different periods in the life cycle on mammary tumorigenesis. The larger sample sizes and longer duration of such studies compared with most basic studies in animal models increase costs and limit their use. However, these studies should be pursued, particularly to explore mechanisms by which these exposures may influence breast cancer cell growth. The underlying mechanisms explaining variable responses to specific exposures associated with familial or genetic predisposition for disease have not yet been explored in animal models with genetic predispositions for obesity, diabetes mellitus or breast cancer, and these approaches may be a promising area for

future research (Yen et al. 1994). In existing epidemiologic studies, these issues can be explored with the addition or analysis of questions pertaining to family history of breast cancer and diabetes mellitus and to personal weight history. These questions should address weight changes over time and events that may be associated with hyperinsulinemia, such as marked weight gain during pregnancy or gestational diabetes. Furthermore, research is needed to identify the active metabolites of androgens and estrogens that promote breast cancer cell growth, particularly to determine whether different metabolites are more active at different periods in a woman's life.

Because of increases in mammography screening for breast cancer, early stage disease represents an increasing proportion of breast cancer incidence. Therefore, it is important that we increase our understanding of factors that improve prognosis as well as factors that prevent initiation of cancer. Three main assumptions have driven the direction of research on initiation. One is that initiation of breast cancer begins in puberty or before; the second is that initiation later in life is less relevant due to the long latency period for cancer; the third is that interfering with the initiation of a normal cell to cancer is more effective in preventing clinical breast cancer than is slowing or stopping the growth of initiated cancer cells. These assumptions have led to a focus on puberty as a critical period in breast cancer prevention in both basic science and epidemiologic research. However, new findings on the possible importance of apoptosis, or programmed cell death, in clinical breast cancer and data suggesting that latency may be shorter than previously estimated suggest that both initiation and promotion may be important in breast cancer prevention throughout life. Epidemiologic research should continue to explore how postulated risk factors influence risk throughout the life cycle. Basic research in animal models has generally been limited to examining the effect of putative risk factors early in life and should be expanded to determine whether exposures have different effects at different periods of the life cycle.

#### *Dr. Birt: Limitations and synergy of epidemiologic and experimental approaches*

Considerable improvement in relating epidemiological and experimental animal data on the relationship of diet to cancer has occurred in recent years. Symposia such as this are certainly helpful. However, interrelating data from these approaches is not integration. Integration can occur only as the questions being asked and the models being used allow parallel investigations.

Investigators who base their hypotheses and experimental approach on both human models and animal models strengthen their research. Studies with humans are certainly more definitive and are clearly necessary to demonstrate that a particular dietary intervention alters human cancer risk. However, intervention trials with humans are limited because usually only one intervention (or a few) can be conducted in a given study. Thus, if the intervention is not successful, the investigator is left wondering whether a slightly different intervention would have had a greater effect. Animal investigations are certainly useful for conducting broader surveys and for identifying potential mechanisms for dietary effects on cancer. But studies with animals are limited in that the diversity of genetic and metabolic backgrounds are not represented by a single animal strain. Furthermore, the animal model will never be as close to the human model as we would desire.

An example of synergy between epidemiological studies and experimental animal research is the interplay in the area of dietary fat and energy in breast cancer. Early investigations

with laboratory animals supported the hypothesis that consumption of high fat diets enhanced breast carcinogenesis (Welsch 1992). Because prospective human trials did not provide evidence for this hypothesis (Goodwin and Boyd 1987, Willett 1990a), animal experiments demonstrated the importance of dietary energy in the enhancement of breast carcinogenesis (Welsch 1992).

Epidemiologists are now challenged to improve data on the relationship of breast cancer to human consumption patterns, physical activity and metabolic profiles, and animal experimentalists are challenged to uncover mechanisms and develop markers of energy intake and utilization.

#### **Dr. Birt: Improvements in animal models**

In the near future, we can expect to see considerable improvements in animal models, especially from the standpoint of genetics. In the past, animal models of breast cancer were believed to be adequate to probe the relationship between diet and cancer if the induced tumors shared the morphology and, to the extent possible, the biology of the human disease. At present, and looking toward the future, it will be more important for animal models to share the genetics of representative human cancers. This will become possible as we understand which genes are important in human cancer. Furthermore, genetic factors in cancer include both patterns of gene expression and the more traditional germ line mutations that predispose to cancer. Nutrition may be important in modulating cancer with both of these genetic influences. Studies of transgenic mice have demonstrated that dietary energy restriction can be as effective in cancer prevention in mice with accelerated tumor development due to *p53* knockout as it is in mice with a wild-type background (Hursting et al. 1994). The study of dietary modulation of different genetic profiles that predispose to cancer is in its infancy. This is an area that will benefit from integration of the experimental and epidemiologic approaches.

#### **Drs. Kestin and King: Controlled dietary interventions and intermediate endpoints**

The major evidence of associations between diet and the risk of breast cancer comes from animal and *in vitro* studies and from epidemiologic studies of human populations. As has been discussed in this symposium, there are serious limitations in present methods of investigation of the role of dietary factors in human breast cancer etiology.

For animal and *in vitro* studies, the applicability to human disease is difficult to assess due to issues such as 1) the choice of experimental animal models (e.g., mammary tumors in rodents may be different from those in humans, being more highly differentiated and less likely to metastasize); 2) the common use of carcinogens in large doses to induce tumors in young animals; and 3) feeding of experimental diets that may not resemble common human diets, such as those that are highly purified or extreme in nutrient intake (Ip 1993). Although much useful data, especially related to mechanisms, has been generated from these studies, we are still left unclear about the relevance to human breast cancer.

Epidemiologic approaches have also been useful in elucidating some of the major risk factors for breast cancer such as age at menarche, parity, and age at menopause. However, despite large numbers of published observational and ecologic studies of diet and breast cancer, there is much controversy about these findings (Hunter and Willett 1993). The lack of consensus derives from measurement issues in dietary assessment.

These issues have been reviewed in detail elsewhere (Willett 1990b); briefly, these studies may be biased by problems such as ascertainment of usual and past diet and confounding of nutrients with each other (low fat diets tend to be higher carbohydrate diets, for example).

A very powerful study design is the randomized, controlled trial. In the area of diet and breast cancer, there is a National Institutes of Health-funded study, currently underway, called the Women's Health Initiative (Women's Health Initiative Clinical Coordinating Center 1994). One of the components of this study is a randomized trial of a low fat dietary intervention to prevent breast cancer in postmenopausal women. However, this study is very resource intensive, involving 48,000 women in 40 clinical centers who are being followed for 9–12 y. Whatever the results of this trial, it cannot, for example, address the question of whether diet may be more important during adolescence, when breast tissue is actively growing (Pike et al. 1993). In addition, it would not be logistically possible or scientifically rational to perform randomized prevention trials of all the dietary factors, let alone their interactions, that have been suggested to be involved in breast cancer etiology.

Another model is to perform controlled dietary intervention studies with human participants using intermediate endpoints. In the pathway from ingestion of a dietary factor to development of disease, there are many steps. For example, we could postulate that a high fat diet increases estrogen production in postmenopausal women by increasing adipose tissue mass, which is a major site of estrogen production in postmenopausal women (Pike et al. 1993). Increased serum estrogen concentrations may increase breast cancer risk because of the documented proliferative effect of estrogen on breast ductal epithelium (Pike et al. 1993). Serum estrogen concentration would therefore be the intermediate endpoint.

The advantages of controlled dietary interventions with intermediate endpoints include the fact that these studies can often be performed in a much shorter time (weeks to years) than the many years needed if cancer incidence is the endpoint. Many fewer participants are involved, and because the diet can be much better controlled, the dietary assessment issues that plague observational studies are largely obviated. This allows for many different dietary factors to be tested, including their interaction. This type of research is more common in studies of diet and cardiovascular disease, from which we have obtained very good data on the effect of dietary factors and interactions on blood pressure and serum lipoproteins (NRC 1989). Another advantage is that mechanisms can be investigated, which can be difficult in many epidemiologic studies.

A major potential disadvantage of the use of intermediate endpoints is that there needs to be good evidence of a causal relationship between the intermediate endpoint and the disease in question. Examples of well-accepted intermediate endpoints include blood pressure for cardiovascular disease risk and adenomatous polyps for colon cancer risk.

At present, there are no well-accepted intermediate endpoints for breast cancer risk. There is evidence, for example that high exogenous circulating estrogen concentrations may increase the risk of breast cancer in postmenopausal women (Toniolo et al. 1995), and there have been dietary intervention studies published on the effect of diet on circulating estrogens (Prentice et al. 1990). However, much more evidence of a causal relationship between circulating estrogens and breast cancer is required before its acceptance as an intermediate endpoint. Dietary associations with other endpoints—such as expression of breast ductal fluid and histology of epithelia

cells (Petrakis 1993), and estrogen metabolism (Bradlow et al. 1995)—are currently under active investigation. Other potential endpoints include proliferation of ductal epithelium, mammographic density, incidence of some forms of benign breast disease, and DNA damage or repair.

Controlled dietary studies in human participants hold great promise in bridging the gap between animal and *in vitro* studies and epidemiologic studies and in elucidating the relationship between dietary factors and breast cancer. These types of studies also provide insights into mechanisms. However, the full realization of the potential of controlled dietary studies awaits the development of well-accepted intermediate endpoints.

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