

Sister Study Hopes to Answer Breast Cancer Questions

Working out of their office in Research Triangle Park, North Carolina, a pair of NIEHS researchers are laying the groundwork for what may prove to be a landmark study of the possible interplay between genetics and the environment in the development of breast cancer. Principal investigator Dale Sandler, acting chief of the institute's Epidemiology Branch, and colleague Clarice Weinberg, chief of the Biostatistics Branch, plan to recruit 50,000 female volunteers aged 35–70 whose sisters have been diagnosed with breast cancer and follow their health over a period of time. Eventually the researchers will evaluate the 1,500 or so of the women that they estimate will develop breast cancer during the initial five years of follow-up, analyzing environmental, genetic, and health data captured at the outset of the study.

Sandler and Weinberg are absorbed in the extensive work necessary to develop a study of this size, scope, and importance. Besides developing an overall plan for the study, the researchers are meeting with focus groups, doing feasibility studies, developing applications and questionnaires, and marshaling the considerable technical, financial, and professional resources necessary to undertake the study, which will run at least 10 years.

"Despite great interest in trying to discover environmental causes of breast cancer, we don't have a lot of evidence," Sandler says. "We don't have a lot of clues; there are no obvious places to start." Some evidence suggests that pesticides and solvents may cause breast cancer. Electromagnetic fields have been the focus of other studies, though Sandler cautions that evidence from those studies is inconsistent. Phthalates, compounds used as plasticizers and fixatives and found in diverse products including cosmetics and lotions, may be a culprit as well. A recent study by the Centers for Disease Control

and Prevention that measured phthalate concentrations in urine samples found evidence of widespread exposure. The highest levels of exposure to monobutyl phthalate, a reproductive toxicant in animals, were found in women of reproductive age. Phthalates are classified among chemicals thought to disrupt reproductive hormones. From that information, the scientists suspect a breast cancer link, too.

Sandler says retrospective and case-control studies such as the Long Island Breast Cancer Study Project and the Carolina Breast Cancer Study have the advantage of including large numbers of breast cancer cases, but because they are retrospective, they may not accurately characterize levels of exposure prior to the onset of breast cancer. Instead, she and Weinberg believe a prospective study will be better able to address factors that cannot easily be measured retrospectively or that may change once a woman develops breast cancer.

Besides collecting biologic and environmental samples such as blood, urine, tap water, and household dust from subjects at

the outset of the study, researchers will also use questionnaires to gather a multitude of data about health history, past environmental exposures, and lifestyle. Sandler believes analyses based on samples collected well before the onset of disease will yield more accurate data for at least two reasons. "One," she explains, "women's exposure patterns could change once they are sick, and if you are measuring something that reflects recent exposures, like diet in the last twenty-four hours, what you get may not represent usual exposure levels. Two, the illness itself or its treatment may lead to changes in the levels. . . . If you are measuring something intrinsic [for example, hormones or immune markers], you would not know if the levels were high because the women were sick or if high levels in the past caused the women to become sick."

To execute such an ambitious study, the research team will need large numbers of participants. Though Sandler and Weinberg expect recruiting the large cohort necessary to generate a statistically viable group to be a challenge, they plan to surmount this hurdle by going after precisely the participants with the most motivation to enroll and stay enrolled—women who have a sister with breast cancer.

"First-degree relatives, especially sisters, have about two times the risk of developing breast cancer as the average woman," Sandler says. Sisters of breast cancer patients are also likely to be within the same age range (and therefore at an age when they are at risk for developing breast cancer) and likely to have been exposed to the same environmental risk factors during early childhood. And of course they share many of the same genes. Sandler says sisters are also likely to share genetic polymorphisms that might affect the way a person's system handles carcinogens or repairs DNA.

The study should also fuel rewarding professional collaborations within the NIEHS, the NIH, and eventually with extramural colleagues too. "We're already building partners [such as the National Cancer Institute] into the study's design," Sandler asserts.

In focus groups of women of diverse socioeconomic, racial, and lifestyle backgrounds, Sandler and Weinberg have discovered that the women have



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one thing in common: as sisters of breast cancer patients, they share a common concern over the disease. That, says Sandler, makes them more likely to want to participate in the study and more willing to comply with the demands of the study, which will lead to a better response rate and higher retention rate over time.

The researchers are building a Web site for the project, developing a marketing plan to attract study participants (including advertisements in women's magazines and cancer publications), finalizing questionnaires, and securing preliminary scientific reviews to validate the integrity of the study's design. "We also have to work things out like standardized blood sampling," Weinberg adds.

"We're hoping by the first of next year to phase in the study," Sandler says. "We'll go into three or four test cities to try the marketing plan and see if it gets women to respond. Then we'll work on getting the bugs out and assess what we need to do to get an ethnically diverse group of women," she adds. In this first phase, 2000 women will be enrolled in the study. Sandler and Weinberg expect it to take three years to enroll all 50,000 participants.

Women enrolled in the study will provide periodic updates during the study and will be given follow-up questionnaires to account, for example, for diet changes or menopause. "By starting with a cohort of women and following them over time, we'll be able to look at many conditions of concern to women in addition to breast cancer, like osteoporosis and thyroid and autoimmune disorders," Sandler says. "This [database of study information] will be a rich resource for lots of nested studies," she adds, "and for addressing new questions that come up over time."

Over time, the study cohort's value to the researchers will increase. "If we do the study for five years, we'll have enough [breast cancer] cases to look at the interaction between many of the exposures of interest and the genes of interest," Sandler explains. "Going ten years for rare exposures or rare genes . . . will give us even more statistical power."
—Jennifer Medlin

the
Sister Study

For more information, check out
<http://www.4sister.org/>
or contact 1-877-4SISTER

NTP Announces Bioassay Results

On 3 May 2001, the National Toxicology Program (NTP) presented for public review five technical reports by the NTP's Board of Scientific Counselors Technical Reports Subcommittee at the NIEHS. NTP technical reports describe an ongoing series of toxicity and carcinogenicity studies conducted on chemical or physical agents to which humans are exposed. The findings from these studies are unique information and knowledge used by health and regulatory agencies in making decisions that affect public health.

Each report reviewed at the meeting involved a series of short- and long-term studies in which male and female rats and/or mice were given a range of doses of test chemical followed by extensive histopathologic examination plus characterizations of clinical chemistry, metabolism and disposition, and genotoxicity. The NTP applies established criteria to classify the strength of the experimental evidence for carcinogenicity in these studies, and the board's subcommittee review included a rigorous evaluation of the studies' findings.

Acrylonitrile and methacrylonitrile—Over 1 million tons of acrylonitrile are used each year in the production of acrylic fibers, elastomers, plastics, and resins. Acrylonitrile is known to be a multisite carcinogen in rats and has been classified as a likely human carcinogen. The NTP studied the effects of acrylonitrile given to mice by gavage and found clear evidence of carcinogenic activity in the forestomach and harderian gland of the eye for both males and females.

Methacrylonitrile is used as an alternative or replacement for acrylonitrile in some of the same applications. No evidence of carcinogenicity was seen for methacrylonitrile in male or female rats or mice. Methacrylonitrile also tested negative in a battery of genetic toxicity assays.

***o*- and *p*-Nitrotoluene**—The *ortho*- and *para*- isomers of nitrotoluene are widely used in production of agricultural and rubber chemicals and a variety of dyes for cotton, wool, silk, leather, and paper. *o*-Nitrotoluene yielded clear evidence of carcinogenic activity in all four sex/species groups, producing skin and mammary gland neoplasms in male and female rats and hemangiosarcomas and large intestine carcinomas in male and female mice. In supplementary mechanistic studies, NIEHS scientists identified *p53* and β -catenin gene mutations in hemangiosarcomas taken from mice exposed to *o*-nitrotoluene; similar mutations were rarely seen in spontaneously occurring hemangiosarcomas from control animals.

By contrast, *p*-nitrotoluene yielded some evidence of carcinogenic activity in female rats (clitoral gland neoplasms) and equivocal evidence of carcinogenic activity in male rats and male mice.

Citral—Citral is a lemon flavoring and fragrance used widely in foods, cosmetics, and other consumer products. Because of the volatility and reactivity of the chemical, it was administered inside starch microcapsules mixed with the animals' feed. This was the NTP's first report using this route of administration in long-term studies. A marginal increase in malignant lymphomas in female mice was judged to be equivocal evidence of carcinogenic activity; there was no evidence of carcinogenicity of citral in rats or in male mice.

A listing of all NTP technical reports including an abstract of each study is available from the NTP home page, located at <http://ntp-server.niehs.nih.gov/> (see NTP Study Information). The Environmental Health Information Service (EHIS) also maintains a library of NTP technical reports, adding new reports as available. Access to the EHIS is available online at <http://ehis.niehs.nih.gov/> or by calling 1-800-315-3010. —Mary S. Wolfe

