Current Research and Future Needs

Contents

Boxes

Tables

Current Research and Future Needs

Since 1983, the U.S. Public Health Service (PHS) has been working to outline an agenda for women's health. In that year, the Assistant Secretary for Health commissioned the PHS to form the Task Force on Women's Health Issues to "identify those women's health issues that are important in our society today and to lay out a blueprint for meshing those issues with the priorities of the Public Health Service" (60). In the ensuing years, controversy over the adequacy of the Federal response to the health service and health science needs of American women has persisted; debates range from whether enough research dollars are allocated to women's health issues, to whether women are sufficiently represented in clinical trials, to whether treatment of disease varies (inequitably) depending on the sex of the patient.

In response to the controversy about attention to research on women's health, the research activities supported by the National Institutes of Health (NIH) were inventoried by the General Accounting Office (GAO), based on finds expended in 1987, to determine resource allocation according to gender. The inventory used the following criteria to identify health problems:

- diseases or conditions that are *unique* to women or to some subgroup of women,
- diseases or conditions that are *more prevalent* among women,
- diseases or conditions that are *more serious* for women or for some subgroup of women,
- diseases or conditions for which the *risk factors are* different for women or for some subgroup of women, and
- diseases or conditions for which the *interventions are* different for women or for some subgroup of women (30).

The inventory determined that 13.5 percent of the NIH budget for fiscal year 1987—approximately $778 million-was spent on women's health issues (30). Certain groups interpreted this figure to mean that, conversely, 86.5 percent of NIH funds went for research focusing on men's health. NIH responded that 80 percent of all its research funds were allocated "either for studies of diseases which affect both men and women or for fundamental research

which has significance for all segments of our population" (30). The inventory did not address whether such research included women in the study populations or used female animal models.

The issue of including women in NIH-funded research study populations has generated growing concern among members of Congress, many of whom regard NIH's response to the matter as inadequate. Congress, therefore, requested GAO to review NIH policy and practices in this area. GAO reported that NIH had "made little progress in implementing its policy to encourage the inclusion of women in research study populations" (58). NIH's response to this criticism was that, "[if] the GAO had done a complete examination of all the data, it would have found that, in the vast majority of research studies, clinical trials and large-scale studies, women are well represented in the study population" (30). On August 24, 1990, in the *NIH Guide for Grants and Contracts, the* agency published a revised, strengthened announcement regarding the inclusion of women in clinical studies. This expanded *Policy Notice* was republished on February 8, 1991, and will be reprinted twice a year.

In reaction to the controversy and concern produced in Congress by the GAO report, the NIH Office of Research on Women's Health was estab-

Photo credit: National Institutes of Health

Building 1, National Institutes of Health, site of the Office for Women's Health Research.

lished in September 1990 to strengthen and enhance NIH efforts to improve the prevention, diagnosis, and treatment of illness in women and to enhance research related to diseases and conditions that affect women. This office, and the Office of Science of the Alcohol, Drug Abuse, and Mental Health Aminstration (ADAMHA), provided the Federal funding figures for research on the menopause and related topics that are presented in this chapter. In addition to presenting these data, the chapter discusses methodologic considerations relevant to studies of the menopause and its treatment, describes completed and ongoing menopause-related studies, and suggests areas for future research.

CURRENT STATE OF RESEARCH ON THE MENOPAUSE AND POSTMENOPAUSAL WOMEN

Previous chapters have referred to the results of studies relevant to the effects of the menopause on the current and future health of women. In addition to these studies, several large-scale investigations with long-term followup involve postmenopausal women. These studies do not necessarily address transmenopausal changes, and hence the impact of the menopause per se, but some of the data they have collected are relevant to these issues. The following section briefly summarizes the objectives and some of the principal findings of these investigations.

The Baltimore Longitudinal Study of Aging

The Baltimore Longitudinal Study of Aging (BLSA), begun in 1958, is an intramural study of the National Institute on Aging to investigate normal human aging. The participants are highly motivated; they are self-recruited, mostly white, educated professionals who return every 2 years for 2 1/2 days of medical, physiological, nutritional, and psychological evaluation (48). The women's cohort was added in 1978. Currently, 441 women, ranging from 20 to 97 years of age, participate in this ongoing observational study. One of its major objectives is to identify and characterize cross-sectional and longitudinal age-related changes among a host of physiological, behavioral, and psychological variables observed in a healthy, community-dwelling population. A second objective is to elucidate risk factors for particular age-related diseases.

Preliminary findings of the BLSA have included the observation that women before the

menopause have a lower incidence of hypertension and heart disease. In addition, diagnoses of cardiovascular disease differ between the sexes: women present for treatment with more angina than men but with fewer myocardial infarctions. More men than women are likely to undergo surgery to remedy their cardiovascular problem.

Physical fitness, measured as maximal oxygen consumption during treadmill testing, is less in women than in men (in large part because women have a lower level of muscle mass) and declines with age in both sexes. Although women have a greater percentage of body fat than men, they also have a lower waist-to-hip ratio (WHR) in their distribution of fat (which suggests a reduced risk of hypertension, diabetes, cardiovascular disease, and mortality compared with men, who have a higher WHR). Although a surge in WHR occurs in postmenopausal women, at any given age, women still have a lower fat distribution ratio than men. The full significance of the WHR is not understood, however, and the meaning of the difference between men and women on this measure is even less clear (57).

Given its broad-based multidisciplinary approach and capacity for longitudinal followup, the BLSA could conduct highly cost-effective research into the effects of the transition through the menopause on many important biological and environmental factors. Ultimately, these data could be used to generate information about perimenopausal risk factors relevant to morbidity in women. However, the inclusion in the study of only approximately 100 women between the ages of 40 and 60 limits its ability to distinguish the effects of the menopause from longitudinal changes or to describe perimenopausal attributes.

The Framingham Study

The Framingham Study began in 1948 as a prospective long-term surveillance study to identify factors related to sex, heredity, and environmental variables (e.g., educational and occupational status, physical activity, smoking) that might be involved in the development of atherosclerotic and hypertensive heart disease. The effort is funded by the National Heart, Lung, and Blood Institute of NIH.

The study recruited two-thirds of the population of Framingham, MA (with the later addition of 740 volunteers), as participants; of these individuals, who were all between the ages of 30 and 59, 2,873

were women. This cohort was offered a physical exam, a standardized cardiovascular exam, and a chest x-ray every 2 years. Medical and family histories were taken, and samples were obtained to determine blood glucose, cholesterol, and hematocrit; urinalysis was performed for other chemistries. Endpoints, or indicators, of coronary artery and cerebrovascular morbidity and associated mortality were evaluated.

The study produced several interesting menopause-related findings. Menopause in this population occurred about a year earlier in smokers (at 49.3 years) than in nonsmokers (50.1 years). Transition through the menopause (naturally or as a result of bilateral oophorectomy) was associated with a rise in levels of hemoglobin and serum cholesterol; it was not associated with changes in body weight, blood pressure, glucose, or vital capacity (22). The study also found an increase in the incidence of coronary heart disease among postmenopausal women compared with premenopausal women, as well as an increase in the severity of the disease when women came to treatment. Surgical menopause was associated with a 2.7-fold increase in the risk of coronary heart disease (19). A particularly important finding was that estrogen users who were also smokers had an increased risk of myocardial infarction (65). Moreover, contrary to most studies of users of estrogen, the Framingham researchers found a 50 percent increase in the risk of cardiovascular disease and a doubling of the risk of cerebrovascular disease in estrogen users compared with nonusers. This result occurred despite the more favorable lipoprotein profiles (higher HDL [high-density lipoprotein] cholesterol and lower LDL [low-density lipoprotein] cholesterol) of estrogen users.

Further consideration of the Framingham findings has produced some reevaluations of these conclusions, however. A reanalysis of the data from the study has revealed that the apparent elevation in risk was not significantly different when only myocardial infarction was considered; in addition, the findings concerning overall elevation in risk were sensitive to the investigator's choice of baseline risk data (52). Moreover, there were only 302 estrogen users in the cohort (51).

In a recent study of offspring of participants in the original Framingham research, postmenopausal estrogen users were shown to have higher levels of the principal protein constituent of HDL, lower LDL cholesterol and glucose levels, and lower diastolic blood pressure than nonusers. Estrogen use was associated with higher levels of HDL cholesterol, but only in women who had undergone oophorectomies (10).

In the original study population, the form of hormone therapy was oral conjugated estrogens (usually Premarin). In the offspring study, Premarin again was the principal therapy used, but 10 percent of hormone users were also taking progestins (which were not identified). Neither study, therefore, provides data regarding the effect of progestins on cardiovascular disease.

The Healthy Women Study

The Healthy Women Study was begun in 1983 and originally consisted of a cohort of 541 Pittsburgh, PA, women who were premenopausal and between the ages of 42 and 50 (36). It was funded, in part, by the National Heart, Lung, and Blood Institute of NIH. The objective of this 5-year prospective longitudinal study was to document normal transmenopausal changes in biological and behavioral characteristics, especially those relating to cardiovascular disease. The study excluded women who had hypertension, thyroid disease, or diabetes mellitus, as well as those who had undergone hysterectomy or bilateral oophorectomy. Medications that might alter the risk factors under investigation were not permitted. Researchers evaluated blood pressure and collected specimens to assess menopausal status, related hormones, and glucose and lipid metabolism. They also assessed medical histories, body size and shape variables, and lifestyle variables such as dietary intake, physical activity, and smoking history.

Although a natural transition through the menopause appeared to have no effect on blood pressure, glucose metabolism, or caloric consumption or expenditure, it was associated with an increase in LDL cholesterol and a decline in HDL cholesterol (36). In addition, declining estrogen levels during the perimenopause were significantly related to the worsening lipid profile (32). Weight gain, a common menopausal occurrence, was significantly associated with a worsening of cardiovascular risk factors such as increases in blood pressure, total and LDL cholesterol, triglycerides, and fasting insulin (66). Compared with

nonusers, postmenopausal women who used estrogen (with or without the progestin medroxyprogesterone acetate) had lipoprotein and cholesterol profiles compatible with a reduced risk of atherogenesis. However, estrogen users also had elevated systolic blood pressure and elevated triglyceride levels (16).

Analyses of the data from this 5-year study are continuing to appear and will provide much needed insight into the natural history of the menopause with respect to the effects of changes in ovarian hormone levels on risk factors for diabetes and cardiovascular disease. Because regimens of hormone therapy are self-selected and not randomized, however, caution is advisable in interpreting the effects of such therapy on measured outcomes. Furthermore, continued followup is critical to ident@ possible links between changes attributable to the menopause and long-term endpoints of morbidity and mortality.

The Leisure World Study

This prospective study of 8,881 postmenopausal female residents of a retirement community in southern California evaluated the use of estrogen in terms of overall mortality (21). The women were almost entirely Caucasian, moderately affluent, and well educated. Their median age at the time of study was 73 years. After 71/2 years of following the women, there had been 1,447 deaths. Women with a history of estrogen use had an age-adjusted, all-cause mortality rate that was 20 percent lower than that of lifetime nonusers. Mortality decreased with increasing duration of use and was lower among current users than among women who had used estrogen only in the distant past. Current users with more than 15 years of estrogen use had a 40 percent reduction in overall mortality. Among oral estrogen users, relative risks of death could not be distinguished by specific dosages of the oral estrogen taken for the longest time.

Women who had used estrogen showed reduced mortality from all categories of acute and chronic arteriosclerotic disease and cerebrovascular disease, compared with nonusers. This group of women also had a reduced rate of mortality from cancer, although this reduction was not statistically significant. Mortality from all remaining causes combined was the same for estrogen users and lifetime nonusers.

The Lipid Research Clinic Mortality *Followup* Study

The Lipid Research Clinics (LRC) Prevalence Study was a cross-sectional study conducted at 10 North American clinics between 1971 and 1976; it was funded, in part, by the National Heart, Lung, and Blood Institute of NIH. Its objective was to describe the distribution of plasma lipids and lipoproteins within populations of American men and women to determine the relationship between hyperlipoproteinemia and cardiovascular disease. A subset of the population was selected for followup in the Lipid Research Clinic Mortality Followup Study, which provided information on the associations between the reported use of estrogen and subsequent cardiovascular and all-cause mortality. The study's female cohort consisted of 2,270 women (40 percent of whom were selected on the basis of elevated lipid levels) aged 40 to 69. The women underwent assessments of blood pressure, blood chemistries including lipid profiles, evaluations of body weight and size variables, electrocardiograms, and 24-hour dietary recalls. To relate environmental and physiological variables to cardiovascular disease, researchers administered questionnaires on lifestyles, menstrual history, reproductive status, and medication use.

After following the cohort for nearly 51/2 years, investigators found that overall mortality was lower (60 percent less) among estrogen users compared with nonusers (7). In addition, when they assessed the relationship between mortality from all causes and estrogen use with respect to the status of pelvic surgery, they found that compared with nonusers, estrogen users who were bilaterally oophorectomized had the greatest reduction of risk (nearly 90 percent less) followed by hysterectomized women (66 percent less) and gynecologically intact menopausal women (46 percent less).

In a later LRC report after 81/2 years of followup (8), a large portion of the 66 percent reduction in risk of mortality from cardiovascular disease could be explained (statistically) by the elevation levels of HDL cholesterol. In addition to higher HDL cholesterol levels, users of estrogen also had decreased levels of LDL cholesterol.

Additional findings in women (that are not strictly related to the menopause or to hormone therapy) included the absence of a cross-sectional relation-

ship between serum cholesterol and either dietary cholesterol or the ratio of dietary polyunsaturated to saturated fats. The LRC data support the conclusion that high levels of HDL cholesterol constitute a strong, independent protective factor—HDL cholesterol 1evels being an important known lipid-related risk factor for cardiovascular disease in women. Although LDL cholesterol levels in women who experience a natural menopause or bilateral oophorectomy may be elevated compared with levels in premenopausal women, the relationship of this variable to cardiovascular disease in women appears to be less consistent. Among women in the study, LRC findings also showed the well-known positive relationships among smoking, obesity, and cardiovascular disease, and the apparent cardioprotective effect of limited (moderate) alcohol consumption.

Several features of the LRC study warrant careful consideration when interpreting its results. The use of estrogen in the study was self-selected rather than randomly assigned, and reports of the study do not indicate the reasons for its use or nonuse. Although the results of the study addressed the issue of potential selection bias, confounding as a result of self-selection cannot be ruled out. Because most of the women were taking oral estrogens, the findings do not apply to nonoral estrogens or to estrogens combined with progestins. Moreover, dose-response relationships were not evaluated, nor was the effect of duration of estrogen use assessed. Although the use of estrogen is associated with an overall favorable lipoprotein pattern, estrogen users in the study had substantially elevated triglyceride levels, implications of which for cardiovascular disease are unclear. Finally, the study did not address the effect of longitudinal changes on biochemical or physiological variables as women traversed the menopause.

The Massachusetts Women's Health Study, Part 2

Part 2 of the Massachusetts Women's Health Study is a 5-year prospective followup study of a cohort of 427 pre- and perimenopausal women; the study, Part 1 of which was begun in 1982, is being supported by a grant from the National Institute on Aging (NIA) of NIH. The cohort was assembled from a previously studied, community-based ran-

dom sample of 2,500 premenopausal Massachusetts women who were believed to be representative of U.S. women in general.

The objectives of this study are to obtain biological, anthropometric, psychosocial, and lifestyle data from women as they traverse the menopause. The study will assess changes in reproductive hormones and related parameters and determine their relationship, if any, to changes in bone mass and cardiovascular risk factors (e.g., lipid and lipoprotein profries). An important goal of the project is to provide valuable basic information on normal changes associated with the transition through the menopause, which are needed to distinguish normal from pathophysiologic changes in reproductive physiology, bone mass, and cardiovascular risk factors. Study designers hope to continue followup beyond the original 5-year term to assess relationships between intermediate outcomes of morbidity and mortality and hypothesized risk factors (37).

The Nurses' Health Study

The Nurses' Health Study is a prospective project based on mailed questionnaires that have been updated at 2-year intervals since 1976 (15). It is supported, in part, by various institutes of NIH. The main objective of the study is to determine the relationships among lifestyle and environmental or exposure variables (e.g., dietary factors, cigarette smoking, use of oral contraceptives and hormone therapy, estrogen in particular) and diabetes, cardiovascular disease, and cancer in women. The initial cohort consisted of approximately 122,000 married female registered nurses who were 30 to 55 years of age and living in 11 States when the study began.

Findings from a recent report on the project show that although postmenopausal women who had used noncontraceptive estrogens in the past had no increased risk of breast cancer, even after more than 10 years of use, the risk of this cancer among current **users increased 40 percent (15). Researchers found no relationship between the risk of breast cancer and dietary fat intake, cigarette smoking, or past use of oral contraceptives; nevertheless, the risk of breast cancer was increased by 50 percent for current users of the**

pill compared with nonusers.[1] A 30 percent greater risk of breast cancer was seen among women who consumed between three and nine drinks of alcohol per week. Family history of breast cancer, early menarche, late age at the birth of a woman's first child, or nulliparity also increased the risk of breast cancer.

With regard to risk deriving from the use of hormones, oral contraceptives were associated with a reduced risk of ovarian and endometial cancer. Not unexpectedly, unopposed postmenopausal estrogens were strongly associated with an increased risk of endometrial cancer.

At the end of the first 4 years of followup, results indicated that the use of estrogen was associated with a reduced risk of cardiovascular morbidity and mortality. Compared with women who had never used estrogen, women who had used estrogen at least once had an age-adjusted risk of coronary disease that was only 50 percent of the expected risk; current users had a risk that was only 30 percent of that expected (15). In a later report based on 6 years of followup, researchers noted that in this cohort of women (which now ranged in age from 36 to 61), a natural menopause did not increase the age-adjusted risk of cardiovascular disease. However, the relative risk more than doubled for bilaterally oophorectomized women if they had never used estrogen. The enhanced risk in oophorectomized women was eliminated by estrogen use (14).

Other variables that increased the risk of cardiovascular morbidity and mortality in women were cigarette smoking, obesity, diabetes, and current but not past oral contraceptive use. Moderate alcohol consumption led to reduced risk of heart disease but more than tripled the risk of subarachnoid hemorrhage.

Assessments of the relationships among hormone therapy and morbidity and mortality are limited to those that consider the use of unopposed conjugated oral estrogens; in fact, 74 percent of estrogen users in the study report the use of Premarin. In addition, the use of estrogen therapy is self-selected rather than randomly assigned. Because data in this study are obtained by questionnaire and because the 30,000 blood samples obtained since 1989 have not yet been evaluated (due to a lack of sufficient funds),

the study is unable as yet to assess longitudinal transmenopausal changes in physiology (13).

In 1991, the Nurses' Health Study reported on 10 years of followup of 48,470 postmenopausal women who did not have a history of cancer or cardiovascular disease when they entered the study. After adjusting for age and other risk factors, the overall risk of major coronary disease in women currently taking estrogen was found to be 44 percent lower than expected (54).

Postmenopausal Estrogen/Progestin Interventions Trial

The Postmenopausal Estrogen/Progestin Interventions (PEPI) Trial is a newly begun double-blind, placebo-controlled clinical trial of oral conjugated estrogens (Premarin), either unopposed or combined with one of two types of progestin (either Provera or micronized progesterone), in postmenopausal women. The study will also examine two schedules of combined therapy with Provera: cyclical Provera (10 mg given for days 1 through 12 of a woman's menstrual cycle) or continuous Provera (2.5 mg given daily every day). The National Heart, Lung, and Blood Institute of NIH administers the study and is the primary funder; co-funders include the National Institute of Child Health and Human Development, the National Institute of Arthritis and Musculoskeletal and Skin Diseases, the National Institute of Diabetes and Digestive and Kidney Diseases, and NIA. The trial is being conducted at seven centers throughout the United States and involves 120 women at each center. Total funding is expected to exceed $10 million.

The primary objective of the trial is to describe the relationships between the various regimens and differences in cardiovascular risk factors related to plasma HDL cholesterol, systolic blood pressure, plasma fibrinogen, and plasma insulin. The project's secondary objective is to assess the effect of these regimens on lipoprotein profiles and metabolism, endometrial changes, bone mass in the lumbar spine and hip, several measures of quality of life, body mass index, renin substrate, plasma renin activity, aldosterone, and a number of factors relating to blood clotting.

---

[1] The increased risk in current users is speculated to be due to an acceleration in the growth and detection of estrogen-dependent tumors in women at risk.

This study will provide important interdisciplinary data and fill key gaps in knowledge of the short-term effects of various hormone therapy regimens on risk factors for diabetes, cardiovascular disease, and osteoporosis. But the limited picture provided by a 3-year study needs to be enhanced by continued long-term followup to determine the effect of these regimens on endpoints related to morbidity and mortality from disease within the cardiovascular and skeletal systems. Another limitation of the study arises from the decision to evaluate only hormone regimens that use conjugated oral estrogens. Transdermal and other nonoral routes of administration of estrogen warrant investigation, given their potential value for hormone therapy pharmacodynamics. There is also a need to understand the effects of nonoral estrogen on risk factors for cardiovascular disease and osteoporosis. A further limitation of the study, according to some critics, is that it is too small to measure an adequate number of endpoints in a reasonable amount of time (51).

The Tremin Trust: An Intergenerational Research Program

The Tremin Trust Research program on Women's Health was initiated in 1934 at the University of Minnesota and continues today at the University of Utah as a nonprofit program supported primarily through grant awards and contributions. The project, originally titled the Menstruation and Reproductive History program, initially sought to determine the magnitude of the variability in the menstrual cycle among women. In 1935, the project began with a pilot study in which participating University of Minnesota women students recorded the onset and cessation of menstrual bleeds in an effort to dispel the common myth that all women menstruate according to the same cycle, i.e., every 28 days.

Between 1934 and 1965, three primary groups of women were enrolled in the program: a 1930s panel of 2,350 women, a 1960s panel of 1,367 women, and an Alaskan panel of 1,000 women, including both Alaskan Native and Caucasian residents. Over the years, a fourth group of women, comprising the daughters and granddaughters of women in the original panels, has been developed. At present, 1,316 women—representing all four groups and ranging in age from the early teens to the mid-nineties remain active as recordkeepers. Of these currently active participants, 852 are menstruating and 464 are nonmenstruating. The study employs as main data-gathering instruments a menstrual calendar card, a medical report form, and a health report form.

The Tremin Trust is currently conducting the Menstrual and Reproductive History (MRH) Followup Study in collaboration with the National Institutes of Environmental Health Sciences to gather health data from approximately 1,000 women who participated in the original MRH research program prior to 1940. **The aim of the study is to examine the link between menstrual and reproductive history and a variety of health outcomes, including longevity. The Midlife Women's Health Study, another effort of the Tremin Trust in collaboration with Pennsylvania State University, is evaluating women between the ages of 35 and 50 who are still menstruating to document the physical and emotional changes they experience as they approach the menopause (50).**

METHODOLOGIC CONSIDERATIONS

Epidemiology is the study of the relationships of various factors that determine the frequency and distribution of disease; its aim is to estimate or closely approximate cause and effect. Studies of the menopause and the onset of age-related disease are necessarily epidemiologic in nature because populations must be studied (or observed) to better understand the associations among symptoms, the menopause, declining hormone levels, and disease. Studies of the effects of hormone therapy can be conducted as trials (which are usually randomized) to determine the safety and efficacy of the treatment (46). Randomized intervention trials and observational studies are both epidemiologic research; both are expensive and time-consuming because of the need to conduct studies that are large enough to have adequate statistical power to distinguish among a variety of potential effects.

In randomized trials, participants are randomly assigned either to a group that will be treated with the factor of interest (in this case, either estrogen therapy (ET) or combined hormone therapy (CHT)) or to a group that will receive placebo treatment. (Sometimes this group receives the "standard" treatment rather than a placebo.) The purpose of randomization is to equalize other causes or correlates of the disease across the treatment groups; it is

particularly valuable for equalizing the distribution of causes that are unknown or not readily measured.

Sometimes randomization is infeasible, unethical, or unnecessary. In the absence of randomized trials, cause and effect can often be established by the results of observational studies, which, taken together, rule out explanations other than causality. Some of these 'other explanations' include chance, selection bias, and information bias (46). A number of ongoing investigations have attempted to evaluate the effects of the menopause and treatment. Most of these studies have been observational and have contributed to a greater understanding of the menopause and the health of women.

Understanding the Effects of the Menopause

It is not possible to use randomized trials to assess the effects of the menopause on the occurrence of disease: a researcher cannot assign the age at which natural menopause will occur in a woman, and it would be unethical to randomly assign women to a group that would undergo surgical menopause merely for the sake of a research protocol. Observational studies are adequate research designs in most instances. A key concern in such studies, though, is the possibility of a biased assessment of the effects of surgical menopause, which could result from the ''selection' of women to this procedure; i.e., women who undergo surgical menopause may be at higher or lower risk for certain diseases.

Observational studies, particularly case-control and followup studies, have provided considerable evidence about the effects of the menopause on a woman's risk for a variety of diseases. The data indicate that natural or surgical menopause at an early age reduces the risk of breast cancer (27) and increases the risk of osteoporosis (38), and that surgical menopause at an early age increases the risk of cardiovascular disease (53). Problems posed by differences between natural and surgical menopause and subsequent risks for disease can be overcome by conducting large studies that have **adequate statistical power to distinguish among the different effects.**

Evaluating the Effects of Hormone Therapy

Unlike research on the menopause per se, assessing the health effects of hormone therapy may require more rigorous research than is possible through observational studies (2,62). Bias may be a difficult, even intractable, problem with observational studies because women often 'select' themselves to receive therapy (or their doctors select them). For example, hormone users may differ from nonusers in terms of factors that influence the risk of acquiring cardiovascular disease but that are unknown or difficult to measure. If the selection factors (i.e., factors that would "cause" a woman to use hormones) are strongly related to the risk of the disease in question, it maybe necessary to conduct trials in which women are assigned randomly to receive hormone therapy or placebo to ensure that the selection factors—and their effects-are equalized between hormone users and nonusers.

The confounding action of such factors is known as selection bias. Because factors responsible for a woman's assignment to a group in a nonrandomized trial influence the results of the trial in ways that cannot be quantified or even identified, researchers cannot be certain that the effects being observed are attributable to treatment or to extraneous or accompanying factors (6). For example, many studies do not provide the reasons for prescribing or not prescribing estrogen for a woman. Because physicians prescribe estrogen most frequently for symptomatic relief, users initially may be physiologically different from nonusers in their susceptibility to symptoms, and this may be related, in turn, to susceptibility to subsequent disease. Alternatively, women who are found in the nonuser category in studies may be intolerant to hormone therapy, and are thus different from users by virtue of physiological factors that influence tolerance.

It has long been suspected that selection bias might be contributing to a "healthy user" effect in hormone therapy studies. Doctors may not prescribe hormones, or may discontinue their use, for women with preexisting illness, women with a family history of contraindications (e.g., cancer or liver disease), or women who develop such contraindications during therapy. This kind of confounding effect could result in an apparent excess of morbidity or mortality in nonusers of hormones, compared with users. A healthy user effect can also occur as a result of better medical care: physicians require hormone users to undergo more frequent physical examinations than nonusers to renew their prescriptions.

In observational studies, selection factors can be taken into account by statistical "control" for

confounding variables such as cigarette smoking, hypertension, and a wide range of other factors that may be related both to hormone use and the risk of disease (54). If statistically controlling for such variables does not alter the results of the study, an absence of bias is assumed. There are circumstances, however, in which statistical control maybe inadequate. Factors generally considered under the headings of "lifestyle" or 'socioeconomic status" may be related to the risk of particular diseases, and they may also be strongly related to the decision to commence or adhere to hormone therapy. But it may be difficult or impossible to control for the effects of these factors because they cannot be measured with adequate precision. In that case, it may be possible to achieve adequate control only by means of randomization.

For many years, these methodological uncertainties have led to doubts about the existence of a relationship between cardiovascular disease and estrogen therapy. In 1991, however, the Nurses' Health Study (54) addressed many of these concerns about the effects of selection bias. The study design controlled for the various confounding variables, and the results of the study demonstrated an association between the use of estrogen and reduced risk of coronary artery disease.

Controlled, randomized prospective trials can overcome the confounding effects of selection bias by randomly assigning participants to experimental groups. This kind of study design can produce an unbiased assessment of the effects of hormone therapy vis-à-vis its risks and benefits, but it cannot provide an understanding of the whole scope of risks and benefits. Some effects are simply too uncommon to be assessed, even in a large, randomized trial. In addition, some effects, such as reduced mortality from hip fracture, take a long time to become manifest (51).

With the exception of the Healthy Women's Study, the Massachusetts Women's Health Study, and future findings from PEPI, the preceding large-scale studies provide, at best, quite limited information on normal hormonal and physiological changes in women as they progress through the menopausal years. Other, smaller-scale studies offer little additional help. Moreover, few ongoing studies include the objective of evaluating longitudinal changes (beginning with premenopausal women) to determine the effect of ovarian hormone

deficiency on the endpoints of other diseases (in addition to cardiovascular conditions) whose clinical presentation may be temporally removed from the menopause.

Requirements for a Randomized Trial of Hormone Therapy

Randomized trials to assess the health effects of hormone therapy must be large enough to detect protective effects against cardiovascular disease and osteoporosis (or fractures); they must also be of sufficient duration to detect effects that may occur only after relatively long use. Both the use of unopposed estrogen and combination therapy should be assessed. In addition to cardiovascular disease and osteoporosis and fractures, other outcomes of interest are breast cancer, endometrial and other gynecologic cancers, and cerebrovascular disease. Also worth studying (from the risk side of the benefit-risk ratio) is hysterectomy and the morbidity associated with this procedure: concerns have been raised that women receiving hormone therapy are more likely to undergo hysterectomy, possibly because bleeding is a side effect of use (2,62).

Trials should be designed to assess both incidence and mortality. Comparisons of total morbidity and mortality among the treatment groups are important for determiningg overall risks and benefits, but an ''overall' comparison is insufficient to better understand individual risks. For example, researchers should compare effects among various age groups because the benefit-risk ratio may be different at different ages. For hormone therapy, the major benefits may be seen among older women, whereas the major risks may be borne by younger ones; such an outcome would influence the interpretation of the benefit-risk ratio for hormone therapy. The costs of the drug and medical care for each treatment group should also be considered in the comparison of benefits and risks (45).

It will be desirable to continue following participants after a trial ends because the adverse effects of hormone therapy on the risk of breast cancer may not become apparent for a number of years. Randomized trials of hormone therapy will be neither easy to conduct nor inexpensive. For both cardiovascular disease and osteoporosis, the protective effect of estrogen therapy appears to be related to the duration of use: women may have to take the drugs

for relatively long periods (10 to 15 years) before a protective effect becomes apparent. Hormone therapy, particularly combination therapy, can have unpleasant side effects. To be successful, a trial to detect the effects of long durations of hormone therapy use must include major efforts to encourage adherence to therapeutic regimens (45).

The Women's Health Initiative Trial

The Women's Health Initiative intervention trial, proposed by NIH, includes a randomized trial of hormone therapy, currently in the planning stages. The trial will assess not only hormone therapy but the effects of a low-fat diet and calcium/vitamin D supplements as well. It is fashioned as a '3 x 2 x 2' factorial design in which women who agree to participate will be randomized first to one of three groups: estrogen alone, a progestin and an estrogen, or a placebo. Each of these groups will then be divided into a low-fat diet or a no-diet group (creating six study groups). These six study groups will each be randomized once again-into calcium/vitamin D-supplementation or no-supplementation groups. (The scientific justification for trials of the low-fat diet and of calcium/vitamin D supplements has been questioned by some epidemiologists (45,51). For purposes of the present discussion, however, the assumption is made that these interventions are justified.)

The designers of the trial believe that a factorial design is more "efficient" than separate trials, because fewer women are needed than if each treatment were being tested in a separate trial. This argument would be persuasive if the treatments had few and minor side effects and if they were simple to administer (e.g., a single pill taken daily). For example, in the Physician's Health Study, which used a "2 x 2" factorial design in which men were assigned to aspirin only, beta carotene only, both, or placebo, there was a' 'run-in' phase before the trial to screen out men who had serious side effects; in addition, each of the men took only a single pill per day (55). In the Women's Health Initiative trial, however, one treatment does not meet the requirement of having few and minor side effects, and another is not simple to administer: hormone therapy—particularly combination therapy-has common and serious side effects (e.g., bleeding) that discourage adherence, and the low-fat diet requires major changes in the choice and preparation of foods. Good adherence to these treatments will

require a serious commitment on the part of the participants over along period of time (9 years is the planned length of the trial).

No **feasibility studies have been conducted to test whether women will adhere to hormone therapy coupled with any of the other treatment conditions (i.e., low-fat diet, calcium/vitamin D supplementation), nor are there plans to carry out such investigations. Yet several sources suggest that adherence over the long run may be a problem. These include data from observational studies on the proportion of women who have ever used hormone therapy and who use it for long periods (2); experience with adherence problems in the PEPI trial, a randomized trial of hormone therapy (2); and results from a feasibility study of adherence to a low-fat diet (21). It is reasonable to predict that adherence to multiple treatments will be poor, particularly over a period of 9 years. Consequently, the likelihood is high that the trial, as designed, will fail to detect treatment effects, even if they exist (45).**

FILLING THE RESEARCH GAPS

Only in the past few decades have researchers begun to understand the potential effects of ovarian hormone levels on morbidity and mortality, effects that appear to be of even greater import in the light of increasing life expectancies for U.S. women. A hormonal component is now believed to be involved in the etiology of osteoporosis and probably in the cardiovascular diseases as well. Metabolic alterations are possible during the perimenopause and may occur in very different biological systems.

The implications of short-term menopausal symptoms for subsequent pathophysiology, as well as the effects of the symptoms themselves, have never been studied. Undoubtedly, the absence of an understanding of ovarian hormone action and of the effects of ovarian hormone levels on nonreproductive target tissues has severely constrained the generation of hypotheses. A further complicating factor is the marked differences among women with respect to the manifestation of menopausal symptoms and susceptibility to chronic diseases. A major challenge in research to develop strategies to prevent disease and maintain the health of older women lies in exploring the consequences of acute, short-term symptoms, as well as the effect of long-term reductions in ovarian hormones on the development

of disease—particularly conditions that have a long latency period or that are temporally removed from the menopause.

As the proportion of older women in the population continues to grow, the need to focus on the prevention of morbidity and disability in this group increases as well. Prevention will require an understanding of the potential effect of modifying lifestyle variables (e.g., nutrition, exercise, smoking) and the identification and use of appropriate intervention strategies (both hormonal and nonhormonal). Determining appropriate strategies requires substantially improved knowledge of the natural history and sequelae of the menopause, and of the role of exogenous and endogenous estrogens and progestins in the etiology and prevention of disease.

An understanding of the physiological consequences of reduced ovarian hormone levels requires protocols and subject selection procedures that can assess the effects of age and type of menopause (natural or surgically induced by hysterectomy or bilateral oophorectomy) on intermediate biological variables and, ultimately, on the risk of disease. **Sensitivity to the potential roles of age and type of menopause is also important in assessing the effects of hormone therapy because there may be marked differences between younger and older women in tissue-specific responses to therapy and in the benefits to be realized in oophorectomized women compared with those who experience a natural menopause. Other important covariates include the time elapsed since the menopause before commencement of hormone therapy, as well as a woman's history of prior use of hormones.**

Biological Systems That Deserve Special Attention

The Ovary

Normal variations in the cyclical hormone patterns of premenopausal women and age-related changes in the patterns of secretion of the gonadotrophins and of estrogen and progesterone are still incompletely understood. Assessments of postmenopausal ovarian function are needed to compare the risks and benefits of ovarian conservation when hysterectomies are performed. Greater understanding of the reasons for the continued production of estrone (a weaker estrogen), androgens, and testosterone by the ovaries of some postmenopausal women is also required (47). Improved knowledge

of the function of the postmenopausal ovary is essential to determine whether the current surgical practice of removing healthy as well as diseased ovaries during a hysterectomy is warranted. Basic research questions include the following:

- What factors differentiate those women with continued ovarian secretory capability from those without such capability?
- What does a fictional postmenopausal ovary secrete, and how long does it remain functional?
- What are the advantages and disadvantages of continued ovarian production of these steroid hormones or precursors?
- Are there fewer or less severe menopausal symptoms among women with continued ovarian secretion?
- Are rates of bone loss lower in these women?
- Do they have an increased risk of reproductive tissue cancer (increased breast, endometrial, or ovarian cancer)?
- Does a more androgenic ovarian output pose increased risk of cardiovascular disease?

The symptoms of reduced ovarian hormone levels (especially the intensity and frequency of hot flashes) are more severe in oophorectomized women than in women who experience a natural menopause (31); oophorectomized women are also more likely to report depression, loss of libido, and dyspareunia (12). Epidemiological studies show that, compared with women who experience the menopause naturally, oophorectomized women have an increased risk of cardiovascular disease (19,14) and significantly greater rates of bone loss (23). Research that controls for or evaluates the role of age at the time of oophorectomy is critical to an objective assessment of the consequences of oophorectomy compared with those of the menopause. For example, the risk of breast cancer is substantially reduced by premenopausal oophorectomy—the degree of protection being related (inversely) to the age of the woman at the time of surgery (57). To better understand the relationship between current medical practice and long-term health outcomes, the following questions need to be answered:

- How widespread is the practice of prophylactic oophorectomy in pre- and postmenopausal women, and why are there regional differences?

- What are the factors that influence judgments by physicians regarding the necessity of removing healthy ovaries?
- What are the risks and benefits of oophorectomy?
- Can women who undergo oophorectomy be assured that they will be able to tolerate hormone therapy and that it will have beneficial effects comparable to those offered by potentially 5 to 10 years of endogenous gonadal hormones?
- Will long-term use of hormone therapy be required for oophorectomized women, and if so, will this increase the risk of (breast) cancer?

In addition to oophorectomies, hysterectomies and tubal ligations are common surgical procedures among U.S. women. Hysterectomies are performed in many pre- as well as postmenopausal women. Tubal ligations may be performed in relatively young, fertile women to prevent conception. Although data are scarce, this procedure may be associated with gonadal hormone deficiency and such symptoms as dysfunctional uterine bleeding and menorrhagia (1 1).

The effects of hysterectomy and tubal ligation on ovarian function are currently unknown, although studies have shown that hysterectomized women experience more severe menopausal symptoms (44, 49). Do these procedures compromise ovarian function and hasten menopause in premenopausal women? Clearly, more research is necessary to understand the consequences of elective pelvic procedures for ovarian function.

The Breast

Some observational studies suggest that the use of unopposed estrogen or of combined hormone therapy, particularly for many years, may result in small increases in the risk of breast cancer (17,56). As is the case for osteoporosis, bias owing to selection factors is less likely to be a problem in these studies than in studies of cardiovascular disease. Women who are more highly educated are at increased risk of breast cancer, whereas thin women are at lower risk. Thus, potential bias from selection factors is not consistently in one direction, as it is in studies of cardiovascular disease.

Randomized trials that are designed to assess whether hormone therapy increases **the risk of breast cancer cannot be carried out because it is unethical to test a drug that is believed to have either a harmful effect or no effect on the occurrence of the disease in question. Therefore, information about breast cancer from randomized trials must come from trials designed to assess possible protection against other diseases. These** data may be sparse because trials designed on the basis of the sample sizes needed to detect protection against cardiovascular disease and osteoporosis may be too small to detect increases in the risk of breast cancer. In addition, such increases may be related to longer durations of use than are required to reduce the risk of cardiovascular disease or osteoporosis. (For this reason, continued followup after the trial is desirable.) Thus, some information will be available from trials, but the most informative data on the influence of hormone therapy on the risk of breast cancer may come from observational studies (45).

Observational studies (case-control and followup studies) of hormone therapy in relation to breast cancer risk must be large enough to detect relatively small increases in risk that may occur after very long durations of use or well after use is completed. It is particularly important to assess combination therapy because the few data currently available on this regimen raise the concern that it may increase the risk of breast cancer more than unopposed estrogen (17,56). It will also be important to control carefully for the age at menopause: because the risk of breast cancer is less for women who experience an early menopause, and because such women are also likely to use hormone therapy for longer periods than women who experience a later menopause, failure to adequately control for age at menopause could mask a harmful effect of hormone therapy on the risk of breast cancer.

Glucose and Lipid Metabolism

The role of declines in ovarian hormones in age-related increases in insulin resistance and the development of adult-onset or type II diabetes is another important issue that deserves study. Insulin resistance has profound adverse effects on glucose and lipoprotein metabolism and may play a prominent role in atherogenesis and cardiovascular and renal diseases. Yet information is limited both on the effects of transmenopausal hormonal changes and on the influence of exogenous estrogen or progestins (4). There is a critical need for an objective evaluation of short- and long-term effects of pro-

gestins on insulin secretion and action because previously progestins have been associated with the development of insulin resistance and compensatory hyperinsulinemia in experimental studies with rhesus monkeys and in studies of oral contraceptive use in women (3). Most important, although short-term use of progestins may not adversely affect glucose metabolism, after 6 months of use, a progressive hyperglycemia and hyperinsulinemia may occur (39). A recent report showed that glucose tolerance and fasting and 2-hour insulin levels were not adversely affectedl the use of a progestin (Provera) in combination with Premarin, compared with unopposed Premarin; however, the study did not consider duration of use of the combination form (3). The PEPI trials will address some of these concerns.

There is considerable evidence that the responsiveness of glucose and lipid metabolism in women, particularly premenopausal women, to diet, weight loss, and exercise is considerably less than the responsiveness in men of the same age (l). In the case of exercise, responsiveness in postmenopausal women may become more like that in men, implying that endogenous estrogen may act as a physiological buffer, dampening the response of lipids to such factors as diet, weight loss, and exercise. This and many other issues related to lipid metabolism and the effects of postmenopausal hormone therapy have yet to be examined (33). Body fat distribution, which is believed to be important in differentiating lipoprotein risk factors in men and women, may be an important risk factor for men that is not present in women (33,42).

A highly atherogenic form of low-density lipoprotein that appears in increasing quantities after menopause (9) also deserves further investigation, as does lipoprotein (a), which increases throughout the menopausal years (33). The effects of estrogen or progestin on the levels of these lipoproteins are unknown (33).

Cardiovascular System

The bulk of research regarding the role of estrogen in cardiovascular disease has focused on changes in lipid and lipoprotein metabolism. Considerable evidence indicates that an important component of the reported cardioprotective effect of exogenous (unopposed) estrogen is mediated by elevated HDL cholesterol and, to some extent, by reduced LDL cholesterol (34). Because of the ongoing development of different estrogen preparations and routes of

administration, as well as changes in the formulation and scheduling of progestins (which area necessary component of hormone therapy for women with a uterus), the effects of such therapy on lipid metabolism will continue to be an area of active research.

Important variables in natural history studies of the menopause as well as in controlled randomized trials of the use of estrogen and progestin include effects on blood pressure and blood coagulation factors. Another important area of study is the effect of lipoprotein metabolism changes on the vessel wall and on the atherogenic process itself, including effects on cholesterol uptake, vascular reactivity, plaque formation, oxidation of lipoproteins, and local platelet function (33). The long- and short-term effects of progestins on hemostasis and blood pressure are obviously of great concern because thromboembolic and hypertensive episodes were associated with early use of oral contraceptives. The PEPI trial will address some of these concerns.

Other critical areas of study in humans are the assessment of transmenopausal changes and of changes in coronary perfusion, cardiac output, and physical performance that result from hormone therapy. As discussed in box 5-A, the development of animal models—in particular, studies using ovariectomized monkeys-shows great promise for studies of the direct effect of estrogens and progestins on the production and amelioration of human-like atherosclerosis.

Much of the available evidence from observational studies suggests that estrogen users have a reduced risk of cardiovascular morbidity and mortality (17,54); thus, an important issue to be addressed is whether women with preexisting cardiovascular disease will likewise benefit from hormone therapy and in what ways. Also deserving of study is a determination of the effects of hormone therapy among women with known risk factors (e.g., hypertension, hyperlipoproteinemia) for cardiovascular disease.

Randomized trials are needed to determine what proportion, if any, of the observed reduction in risk is due to a real effect of estrogen and what proportion, if any, is due to selective use of estrogen by women already at reduced risk of cardiovascular disease. Such a trial would, of course, also assess the effect of combination therapy. There is as yet no informative evidence on the effect of this drug regimen; because progestins can affect

Box 5-A—Development of Animal Models for the Menopause

Animal models are useful in understanding basic biological mechanisms, formulating hypotheses, and providing evidence of cause-and-effect relationships. It is generally difficult, expensive, or time-consuming to study risk factors in humans because tissues affected by pathophysiological changes may not be readily (or ethically) available and decades may be required for the manifestation of pathophysiological processes. Furthermore, reproducible outcomes, which are fundamental to establishing direct evidence of cause and effect, may be confounded by the severely limited ability to ensure compliance-that is, to control for lifestyle and environmental variables in free-living populations. An additional problem in securing such outcomes is the difficulties that arise in trying to use randomized designs, which prevent bias in the allocation of subjects to treatment regimens.

Although rats have a strain-dependent 4- or 5-day estrus cycle (rather than a 28-day menstrual cycle) and do not experience a menopause, the ovariectomized rat has been proposed and used as an animal model for human postmenopausal osteoporosis, because the loss of ovarian hormones in rats, as in women, results in decreased bone density and more fragile long bones. As in oophorectomized women, ovariectomized rats also have deficits in intestinal calcium absorption, which contribute to the loss of bone mass.

Although it is commonly believed that the ovariectomized rat is not the best animal model because its skeleton continues to grow, this finding is probably not due to any species-specific anomaly but rather to the use of juvenile animals that have not attained skeletal maturity. The selection of a mature rat (before ovariectomy is performed) is essential as a paradigm, because bone loss in postmenopausal women obviously occurs only after the attainment of skeletal maturity. Although the ovariectornized rat is a convenient, practical animal model of bone loss, significant differences in bone morphology with respect to the organizational pattern of skeletal components, compared with humans, limit its widespread use+

Attempts have been made to overcome this drawback as well as to develop an animal model that has a more relevant reproductive physiology and that is susceptible to atherogenesis. Researchers at the Arteriosclerosis Research Center (ARC) and Comparative Medicine Clinical Research Center at Wake Forest University's Bowman Gray School of Medicine (Winston-Salem, NC) have begun evaluating the utility of cynomolgus monkeys as models for menopause-related pathophysiology.

For the past decade, researchers at the ARC have conducted randomized, controlled intervention trials using cynomolgus monkeys to explore the physiological and cellular mechanisms by which estrogens and progestins affect not only bone metabolism and bone density but also coronary artery atherogenesis and vascular tissue responsivity. Because this species of monkey is susceptible to diet-induced atherogenesis, and because female cynomolgus macaques have a reproductive physiology more comparable to that of humans than the reproductive physiology of other primates, these investigators have been able to study directly and simultaneously the role of ovarian hormones on endpoints of both atherogenic and osteoporotic processes. Their investigations have produced valuable insights that would be impossible to achieve from studies of humans.

Scientists from the ARC have provided substantial evidence that the ovariectomized female cynomolgus macaque is a highly appropriate model for the study of menopause-related pathophysiology. The center has studied the effects of ovariectomy and ovarian hormone treatment on bone histomorphometry, bone density, and biochemical parameters of bone metabolism. Researchers have also observed changes in histomorphometric parameters (indicative of a loss of architectural elements) and biochemical markers of bone breakdown that are similar to those seen in postmenopausal women with osteoporosis.

With regard to cardiovascular disease, ARC studies have shown that coronary artery atherosclerosis in female cynomolgus monkeys appears to be morphologically similar to that in women. Furthermore, ovariectomy in this species results in a doubling of the extent of atherosclerosis. Most important, these investigators observed that ovariectomized monkeys given estrogen or estrogen plus progesterone showed a marked inhibition in the progression of atherosclerosis compared with ovariectomized monkeys receiving placebo. It thus appears that the ovariectomized cynomolgus monkey may be a highly appropriate model to study the pathogenesis of age-related diseases in middle-aged and older women.

SOURCES: **Office of** Technology Assessment adapted **from** C. Gennari, D. Agnusdei, P. Nardi, et al., "Estrogen Preserves a Normal Intestinal Responsiveness to l,25-Dihydroxyvitamin D_3 in Oophorectomized Women" *Journal of Clinical Endocrinology and Metabolism 71(5):1288-1293, 1990;* R.P. Heaney, R.R. Reeker, and P.D. Saville, "Menopausal Changes in Bone Remodeling," *Journal of Laboratory and Clinical Medicine 92(6):964-970, 1978;* D.N. Kalu, C.-C. Liu, R.R. Hardin, et al., "The Aged Rat Model of Ovarian Hormone Deficiency Bone Loss," *Endocrinology 124(1):7-16, 1989;* P.D. Saville, "Changes in Skeletal Mass and Fragility With Castration in the Rat: A Model of Osteoporosis,' *Journal of the American Geriatric Society 17(2):155-166, 1969.*

serum lipids adversely, the effect of combination therapy on the risk of cardiovascular disease may not be favorable (17).

Finally, better understanding of the role that estrogen plays in protecting premenopausal women against cardiovascular disease may lead to better diagnosis and treatment of cardiovascular disease in postmenopausal women. For example, some researchers have suggested that the perception that women tolerate cardiovascular disease better than men and hence have a better prognosis may be due in part to the inclusion of women without cardiovascular disease (who are misdiagnosed as having angina) in female patient populations with cardiovascular disease (64). This misunderstanding of the clinical presentation and prognosis of the disease in women may in turn lead to less aggressive preventive interventions (e.g., prescribing therapies, risk factor modification) and the postponement of referral for further noninvasive testing. Because women may be referred for procedures such as coronary bypass surgery later in the course of their disease than are men, they may be older and their condition more serious; not surprisingly, they experience increased operative mortality (29). **As this one example demonstrates, deficiencies in the ability to diagnose cardiovascular disease in women and ultimately deliver better medical care can be corrected only by increased understanding of the etiology and course of cardiovascular disease in this population group.**

Skeletal System

Given the bone status of those women already diagnosed as osteoporotic, it is estimated that half of all Caucasian women will develop vertebral fractures and one-third will suffer hip fractures by the age of 90 (43). Yet at least 50 percent will not have osteoporotic fractures even in extreme old age. Although estrogen deficiency undoubtedly plays a major role in the development of this disease by increasing the rate of bone loss throughout the skeleton, not all perimenopausal women lose bone at the same rate. Furthermore, although enhanced perimenopausal bone loss occurs at all sites, the effect of reduced endogenous estrogen levels on rates of loss varies from site to site; that is, the spine shows the greatest rate of estrogen-sensitive loss compared with the hip and radius, which reflect a greater component attributable to age.

Because estimates of bone loss in women may be based on study populations that commingled oophorectomized women with those who had experienced a natural menopause (not to mention smokers, women who never exercised, and women with a variety of other contributing risk factors), perceptions of what constitutes a "normal" rate of menopausal bone loss may be distorted, leading to overestimates of the number of women who are at risk for osteoporosis. The effects of a natural menopause (and of reduced ovarian hormone levels) need to be evaluated separately and compared with the effects arising from oophorectomy (and from ovarian hormone deficiency) on site-specific rates of bone loss. These studies should also include an assessment of the effect of age at menopause (and years from menopause) on rates of bone loss. Answers to these questions may help distinguish women who lose bone quickly from those who lose bone slowly.

Sensitive, specific metabolic markers are needed to indicate when skeletal depletion is occurring and to help monitor both early and later responses to treatment. Such markers should be capable of quantifying bone resorption and formation and the degree of imbalance between the two (59).

Research is also needed at the cellular level to determine the role of in vivo estrogen deficiency and the effects of estrogen therapy on the factors that regulate the coupling of processes of bone formation with those of bone resorption. Many physicians advocate estrogen therapy as the most effective available treatment for prevention of osteoporosis, but it is not clear how long estrogen (or combined therapy) should be prescribed to prevent fractures in later life. Also unknown is the length of time during which hormone therapy is effective in preventing or reducing rates of bone loss.

Research is needed to answer the following questions:

- Is estrogen therapy as effective in the femur as in the vertebrae?
- What is the effect of long-term progestin use on skeletal mass?
- What is the cumulative effect of periods of starting and stopping estrogen, a practice believed to be common among users of hormone therapy (26,37)?
- Are the effects of estrogen cumulative, or is there a threshold in terms of duration of use? Is

there an acceleration of bone loss when therapy is discontinued?

- Do the benefits to be realized (in bone density maintained or fractures reduced) depend on the age of the patient or the number of years postmenopause?
- Is the initiation of estrogen therapy in postmenopausal older women (over age 65) effective in preventing further bone loss and fractures?

Many observational studies suggest that both the use of unopposed estrogen and of combined therapy reduce the incidence of osteoporosis (or fractures) (17). Bias resulting from selection factors is likely to be less of a problem in these studies than in studies of cardiovascular disease. Thinness is related to an increased risk of osteoporosis and physical activity to a decreased risk. The presence of bias as a result of thin women 'selecting' themselves for hormone therapy will be in the direction of underestimation of a protective effect of therapy, whereas bias owing to the self-selection of physically active women will be in the direction of overestimation. In contrast, bias from selection factors in studies of cardiovascular disease is more consistently in the direction of producing an apparent protective effect of hormone therapy. If the only issue, then, was the effect of hormone therapy on the risk of osteoporosis, randomized trials might not be deemed necessary. However, in the context of trials to assess the effects of hormone therapy on cardiovascular disease, it is worthwhile to assess osteoporosis as well.

Nutrition, Energy Balance, and
Body Composition

With the exception of considerable clinical data on calcium consumption, few studies of middle-aged women have examined the effects of the menopause or of hormone therapy on the requirements for vitamins, minerals, and other nutrients. Alterations in the efficiency of absorption, excretion, and metabolism of various nutrients are known to occur during periods of altered ovarian hormone levels such as those that occur during pregnancy or oral contraceptive use. It was also demonstrated recently that oophorectomy reduces calcium absorption (18). Because it is likely that the efficiency of processes related to nutrient assimilation, utilization, and retention may be compromised or altered by the ovarian hormone deficiency characteristic of the menopausal period, actual nutrient requirements may be different in peri- and postmenopausal women compared with premenopausal women. Considering the potential impact of nutrition on the prevention or modification of disease processes, an assessment of nutrient requirements in menopausal women should be an area of high priority.

Data on the relationships among weight gain and changes in dietary patterns and caloric intake in middle-aged women are also severely limited, particularly with regard to changes in ovarian hormone status. Increases in weight are common in middle-aged women and are strongly associated with enhanced risks of diabetes and cardiovascular disease (66). In fact, an increased risk of cardiovascular morbidity and mortality-which persisted even after controlling for the influence of weight on blood pressure and cholesterol—has recently been demonstrated in overweight, middle-aged women (35).

Although the effects of hormone therapy on body weight in humans have not been systematically explored, it has been reported that estrogen users appear to be significantly slimmer and taller than nonusers at all ages (4). Studies in rodents (63) and nonhuman primates (28) have shown that the gonadal hormones exert profound effects on food intake, weight gain, and body composition. For example, ovariectomy in rodents is associated with overeating, rapid and profound increases in body weight, and obesity. These changes can be prevented or reversed with estrogen treatment. The addition of a progestin, however, counteracts the effect of estrogen and leads, again, to increases in weight and obesity (63). Knowledge of the effects of estrogen, especially in combination with progestins, on the regulation of energy balance and adiposity in middle-aged women is critical to appropriate prescribing of hormone therapy. (See box 5-A for a discussion of animal models.)

Renal Function

Few studies of renal physiology have focused on women in general, let alone on transmenopausal changes or changes as a result of hormone therapy. The kidney plays a major role in mineral homeostasis. Ovarian hormone deficiency and hormone therapy are known to produce marked changes in serum levels and urinary excretion of calcium and phosphorus. An assessment of age- and menopause-related alterations in renal function (particularly in renal tubular function) will foster a better understanding of mineral homeostasis in women of all

ages and provide an additional perspective on peri- and postmenopausal bone loss (47).

A study of water and electrolyte metabolism, with which the kidney is integrally involved in both pre- and postmenopausal women, could lead to better understanding and hence more effective treatment of problems related to water retention and bloating. Such problems, which commonly occur both premenstrually and as an unwanted side effect of hormone therapy (often resulting in reduced compliance with many hormone regimens), can have marked adverse effects on the quality of life of young and old women.

Pharmacology

It is presently assumed that any given pharmacologic agent administered to women will have similar action, metabolism, and disposition, regardless of where a woman maybe in her menstrual cycle or of whether she is having regular cycles. Such an assumption is also made regardless of whether a woman is taking oral contraceptives or is postmenopausal and receiving hormone therapy. This assumption is surprising, given the possibility of profound metabolic alterations (e.g., those reflected in severe vasomotor instability or in changes in bone mineral metabolism) that may result from the menopause, oophorectomy, or hormone therapy. Scant attention has been focused on the issue of whether ovarian hormone status itself or alterations of metabolism secondary to changes in ovarian hormone status may affect pharmacodynamic responses.

Oophorectomized women who are not taking estrogen show a marked reduction in vertebral bone density and a substantial decline in intestinal calcium absorption compared with presurgery baseline values. Oophorectomized women receiving estrogen therapy show no decline in bone mass or calcium absorption (18). This suggests that comparable or presumably adequate blood levels of a given drug or minerals do not ensure comparable pharmacodynarnics in physiological states that are dissimilar because of ovarian hormone status (whether as a result of oral contraceptive use, menopause, or hormone therapy). The question of impaired or altered action of other pharmacologic agents on other tissues as a result of changes in the levels of ovarian hormones calls for critical examination. The efficacy and side effects of drugs should be assessed in populations of women whose ovarian hormone status reflects that of future users of the drugs.

Exercise

Experts increasingly advocate physical activity and exercise as being of benefit to the cardiovascular and skeletal systems and contributing to a sense of overall well-being by their positive effects on mood. Sedentary behaviors and physical inactivity, on the other hand, are associated with a higher incidence of coronary heart disease and with underlying risk factors for atherosclerosis-- e.g., obesity, hypertension, and diabetes (40).

Although physical fitness and cardiovascular disease are not well studied among women, researchers have shown that in comparison with sedentary women, women who regularly engage in endurance activities have higher HDL cholesterol levels (40). Furthermore, women who participate in aerobic conditioning show improvements in glucose tolerance and response to insulin (61). Studies have also found that women who exercise have reduced mortality from cardiovascular disease and cancer, leading to a fourfold reduction in mortality from all causes (5).

To achieve maximal benefits for the skeleton, it is believed that exercise that has a significant weight-bearing component is necessary (41). Although moderate exercise may be effective in increasing or preserving bone mass in the lumbar spine in postmenopausal women, it is unclear whether the beneficial effects of exercise extend to the period of accelerated bone loss during the perimenopause. A number of important questions arise regarding specific recommendations for exercise prescription in women. The effects of exercise on site-specific changes in bone mass, on risk factors for cardiovascular disease, and on long-term endpoints such as fractures and cardiovascular morbidity and mortality all need to be evaluated. In particular, research is needed to establish guidelines on the type, intensity, frequency, and duration of exercise necessary to derive maximal benefits to both the musculoskeletal and cardiovascular systems and to avoid ineffective training and possible injury. In older women, the interaction of exercise with lifestyle or environmental factors such as nutrition or hormone therapy deserves study, because it has been demonstrated among younger women that calcium consumption plays an important role in optimizing the potential benefits of physical exercise on the skeleton (24).

Effects of Ovarian Hormone Levels: Modalities of Study

Given the present dearth of data, both short- and long-term multidisciplinary observational studies could make substantial contributions to an understanding of the natural history of the menopause. Cohorts (of a sufficiently broad age range) of premenopausal and postmenopausal women should be included in initial subject populations to generate immediate cross-sectional comparative data, as well as to establish premenopausal values for the assessment of transmenopausal changes and long-term outcomes. These studies should be multidisciplinary, cutting across the major biological and psychosociocultural systems. They should assess the role of reproductive history vis-à-vis pregnancy and lactation, as well as the effects of *cumulative* exposure to exogenous hormones such as diethylstilbestrol (DES) and oral contraceptives.

There are almost no data on the menopausal experience or on the use of hormone therapy among women of color or women of various ethnic backgrounds. The study of individuals from different genetic backgrounds is valuable not only in understanding unique pathophysiologies but also in understanding differential susceptibilities to disease. For example, compared with Caucasians, African Americans have a substantially lower risk of osteoporosis but a much higher incidence of hypertension, diabetes, and cardiovascular and renal disease. To date, however, research has largely ignored the role played by the menopause and by reduced ovarian hormone levels in these diseases among African Americans and other ethnic subgroups.

With the exception of the PEPI study, there is a decided lack of randomized placebo-controlled or comparable-treatment clinical intervention trials, as well as a lack of trials of clinical endpoints, that address the effects of hormone therapy. Experimental randomized clinical trials of hormone therapy as it affects morbidity and mortality are needed to obtain an unbiased assessment of the effects of such therapy, to develop algorithms regarding prescribing practice, and to assess the effectiveness of nonhormonal interventions.

FEDERAL INVESTMENT IN MENOPAUSAL AND RELATED RESEARCH

In the summer of 1991, OTA requested that NIH and ADAMHA provide budget data on research related to the menopause. The responses of each agency are reported below.

National Institutes of Health

The PHS has no standard definition of hormone therapy that is used throughout the agency. Therefore, NIH chose the following definition, which was provided by the National Institute of Child Health and Human Development, to use in collecting budget data:

> Hormone therapy is the use of exogenous sex hormones for the relief of menopausal symptomatology. The hormones may be synthetic or natural and are derived from humans and animals. The concept of hormone therapy pertains to the use of estrogen and progesterone or synthetic progestin. Estrogen therapy pertains to the use of only estrogen for the same purposes. Hormone regimens pertain to any of the above variations in terms of hormone dosage and duration.

Data were derived from the NIH Computer Retrieval of Information on Scientific Projects (CRISP) database as well as directly from the appropriate institute and center directors (ICDs). CRISP is a major scientific information system containing data on all research programs supported by units within the PHS. Two searches of the CRISP databases were conducted: one based on the single term **menopause,** which included postmenopause, and a second based on the terms **menopause** and *hormone replacement therapy. The* first search revealed $145.5 million in funding (see table 5-l); nearly $72 million of that research is related to breast cancer and is funded through the National Cancer Institute.

The second search revealed no studies specifically associated with hormone therapy; consequently, the search was expanded to include the following related terms: estrogen, estradiol, estriol, estetrol, estrone, estrogen analog, diethylstilbestrol, ethynylestradiol, mestranol, mestranol norethindrone, and mestranol norethynodrel. Including these terms revealed obligated total funding of $15.5 million for fiscal year 1991 (see table 5-2). The figures in the

**Table 5-l—Estimated Volume of General Research
Related tot he Menopause and the Postmenopausal
Period, National Institutes of Health,
Fiscal Year 1991**

Institute, center, or division	Total dollars (in millions)	Total number of projects
National Institute on Aging	$ 5.6	24
National Institute of Arthritis and Musculoskeletal and Skin Diseases	7.8	43
National Cancer Institute	119.7	NA
National Institute of Diabetes and Digestive and Kidney Diseases	3.9	22
National Institute of Child Health and Human Development	1.3	7
National Heart, Lung, and Blood Institute	5.2	21
National Center for Nursing Research	0.4	2
National Center for Research Resources	1.2	52
National Institute of Dental Research	0.4	2
Total	**$145.5**	**173**

NOTE: NA = Data not available.

SOURCE: National Institutes of Health, 1991.

**Table 5-2—Estimated Volume of Research Relatedto
the Menopause and Hormone Therapy, National Insti-
tutes of Health, Fiscal Year 1991**

Institute, center, or division	Total dollars (in millions)	Total number of projects
National Institute on Aging	$ 4.2	15
National Institute of Arthritis and Musculoskeletal and Skin Diseases	1.8	10
National Cancer Institute	1.6	NA
National Institute of Diabetes and Digestive and Kidney Diseases	2.3	14
National Institute of Child Health and Human Development	0.5	4
National Heart, Lung, and Blood Institute	4.4	17
National Center for Nursing Research	0.2	1
National Center for Research Resources	0.2	23
National Institute of Dental Research	0.3	1
Total	**$15.5**	**85**

NOTE: NA = Data not available.

SOURCE: National Institutes of Health, 1991.

table reflect the total funds awarded, including direct and indirect costs.

The data have certain limitations that require consideration. The CRISP database does not distinguish new projects from continuing ones; thus, the dollar amounts do not reflect the total funds spent for research related to the searched term. In addition, a particular project may be counted more than once if it is associated with more than one of the searched index terms.

In total, NIH support for research related to the menopause and its consequences includes studies of basic reproductive, cellular, and molecular biology and more clinically related investigations of, for example, osteoporosis, coronary artery disease, stroke, and cancers of the breast and uterus. In addition, NIH supports research related to changes in various organs and tissues that occur as a result of the physiological changes associated with the cessation of ovarian functions. It also funds efforts that address the resultant symptoms and signs of the menopausal period that concern women and affect their well-being. (See box 5-B for descriptions of the types of research supported by various institutes of NIH.)

Alcohol, Drug Abuse, and Mental Health Administration

ADAMHA, an agency of the PHS, also funds research related to women's health and the menopause. The three ADAMHA institutes-the National Institute on Alcohol Abuse and Alcoholism (NIAAA), the National Institute on Drug Abuse (NIDA), and the National Institute of Mental Health (NIMH)-+onduct and fund research that is both directly and indirectly related to the menopause. Most of the work is done extramurally, but ADAMHA also supports three full-time intramural menopause researchers.

Grant abstracts for the three institutes were analyzed for menopause-related topics. From 1989 to 1991, ADAMHA provided close to $5 million for 32 research grants that were directly related to the menopause (see table 5-3). Many of these projects are oriented toward basic research; only 7 of the 32 grants are for research on human subjects. Those 7 grants fired 3 projects that focus on alcohol effects in postmenopausal women; biobehavioral studies of narcotics abuse, including alcohol-induced changes in endocrine function in postmenopausal women; and research on the psychobiology and treatment of perimenopausal mood disorders.

Box 5-B—Menopause-Related Research at the National Institutes Health

Many **of the institutes, centers, and divisions (ICDs) of the National Institutes of Health contribute resources and support to research on** women's health. Because of the various missions and objectives of the ICIDs, they may **place different emphases on the many areas of womens' health. The following are the primary institutes that provide resources to support research on the menopause and on hormone therapy.**

The National Institute of Child Health and Human Development historically **has been associated with** issues **related to human development and the reproductive health of women. The Center for Population Research leads the** Federal Government's effort in this regard. Through grants and contracts, the center sponsors work ranging from basic biomedical research in the reproductive sciences to epidemiologic studies on the menopause and on the postmenopausal period.

The National Heart, Lung, and Blood Institute (NHLBI) is supporting research that will contribute to an **understanding of how interventions to improve lipid and cholesterol profiles in postmenopausal women may be useful in preventing the progression of coronary artery atherosclerosis. Other research supported by** NHLBI **includes studies on postmenopausal use of estrogen and progestin in relation to the risk of coronary disease.**

The National Cancer Institute **supports studies of postmenopausal women that focus on the prevention, treatment, screening, and detection of cancer and on the relationship of obesity, smoking prevention and cessation, and hormone therapy to the development of malignancy.**

The National Institute on Aging **has begun a major new initiative related to hormonal therapies for osteoporosis, which is reflected in the large increase in funds for the institute in fiscal year 1991. Studies range from longitudinal studies of the menopause, bone loss, and aging to behavioral treatment of menopausal hot flashes.**

The National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK) hasanendocrinology program that includes a significant effort in support of research on the menopause and on hormone therapy. In addition, NIDDK is involved in clinical and basic research on the action of estrogen on various tissues. The institute has a strong research interest in this field, as well as in the closely related field of osteoporosis.

The National Center for Nursing Research supports basic and clinical interdisciplinary research related to women's health across the lifespan.

The National Institute for Dental Research supports research that examines the status of various oral tissues during physiologic aging and, more specific to women's health issues, the observable effects of the menopause and hormone therapy on salivary gland functions.

The National Institute of Arthritis and Musculoskeletal and Skin Diseases provides funds for research efforts on osteoporosis, bone density, and pathological bone reabsorption along with research on the effects of estrogen or combined hormone therapy.

Some studies *supported by the National Center for Research Resources include,* but are not limited to, exercise intervention in relation to bone mineral content in postmenopausal women, ovarian steroids in menopausal women with endometrial cancer, and thermoregulation during menopausal hot flashes.

SOURCE: National Institutes of Health, 1991.

In response to the health promotion and disease prevention goals of the PHS's Healthy People 2000 program, NIMH is seeking to expand its research base on issues related to women's health through basic, clinical, and epidemiologic research. Currently, the agency is designing a program that will encourage extramural research on changes in women's mental health during the lifecycle; one of the research goals of the program is related to changes in mood and behavior that are associated with the menopause.

SUMMARY

As the proportion of older women in the U.S. population continues to grow, the need for prevention or, alternatively, early diagnosis and treatment of morbidity and disability in this group similarly increases. The development of appropriate intervention strategies depends on substantially improved knowledge in two areas: the natural history and sequelae of the menopause and the role of estrogen and progesterone deficiency and replacement in the prevention or modification of disease.

Thus far, hormone therapy is the most efficacious treatment modality for the amelioration of menopausal symptoms and prevention of osteoporosis. Epidemiological and animal studies strongly suggest that estrogen therapy has the potential to reduce morbidity and mortality from cardiovascular disease. But in the absence of randomized clinical trials, a definitive, unbiased assessment of the beneficial effects of estrogen with and without progestin in preventing or ameliorating cardiovascular disease is not possible. Objective evaluation of the risks is likewise precluded. Moreover, there are virtually no studies on the effects of long-term use of estrogen with progestin (i.e., combined hormone therapy)—a recommended treatment regimen for nonhysterectomized women.

Because estrogen and progesterone affect a host of tissues throughout the body, future research should foster an integrated, multidisciplinary approach, such as that used in the new PEPI trial in its multiorgan system evaluation of risk factors and intermediate points of disease. Furthermore, randomized clinical trials of this kind, with their long-term followup studies and assessments of multiple morbidity and mortality endpoints, are crucial to an objective evaluation of risks and benefits. Without such an evaluation, some of the historical disasters that have accompanied the use of exogenous ovarian hormones may be repeated: for example, the increased risk of reproductive cancers in mother and offspring from the use of (or in utero exposure to) DES; the marked number of hypertensive and thromboembolic episodes attributed to use of early formulations of the pill (which took 10 years to uncover, even though the pill was being used by millions of women) (25); and the excess of endometrial cancer in users of unopposed estrogens before the recommendation that progestin should be added.

An increase in the quality of life of older women depends on early improvements in diagnosis, which in turn require the identification of risk factors (that may be different from those for men) and the development of practice plans with regard to preventive and therapeutic strategies. It also requires objective findings from well-controlled studies to determine who will benefit and how, for how long a particular intervention (either hormonal or non-hormonal) will be effective, and in which body systems. It requires increased understanding of women's physiology in general. And it requires the

Table 5-3-Extramural Research Related to the Menopause, Alcohol, Drug Abuse, and Mental Health Administration, 1989-91

Fiscal year	Institute	Institute funding($)	Total ADAMHA funding ($)
1989	National Institute of Alcohol Abuse and Alcoholism	$560,413	
	National Institute on Drug Abuse	480,505	
	National Institute of Mental Health	468,727	$1,509,645
1990	National Institute of Alcohol Abuse and Alcoholism	591,111	
	National Institute on Drug Abuse	432,109	
	National Institute of Mental Health	711,298	1,734,518
1991	National Institute of Alcohol Abuse and Alcoholism	580,374	
	National Institute on Drug Abuse	613,288	
	National Institute of Mental Health	411,865	1,605,527
	3-year total		$4,849,690

SOURCE: Alcohol, Drug Abuse, and Mental Health Administration, 1991.

dissemination of badly needed information regarding the menopause to physicians and their patients.

CHAPTER 5 REFERENCES

1. Barnard, R.J., 'Effects of Life-Style Modification on SerumLipids," *Archives of Internal Medicine 151(7): 1389-1394, 1991.*
2. Barrett-Connor, E., and Bush, T.L., "Estrogen and Coronary Heart Disease in Women," *Journal of the American Medical Association 265:1861-1867,1991.*
3. Barrett-Connor, E., and Laakso, M., ''Ischemic Heart Disease Risk in Postmenopausal Women: Effects of Estrogen Use on Glucose and Insulin Levels," *Arteriosclerosis 10(4):531-534, 1990.*
4. Barrett-Connor, E., Wingard, D.L., and Criqui, M.H., "Postmenopausal Estrogen Use and Heart Disease Risk Factors in the 1980s: Rancho Bernardo, CA, Revisited,' *Journal of the American Medical Association 261(14):2095-2100, 1989.*
5. Blair, S.N., Kohl, H.W., Paffenbarger, R. S., et al., "Physical Fitness and All-Cause Mortality: A Prospective Study of Healthy Men and Women," *Journal of the American Medical Association 262(17): 2395-2401, 1989.*
6. Bush, T.L., "The Epidemiology of Cardiovascular Disease in Postmenopausal Women,' *Annals of the*

New York Academy of Sciences, vol. *592, Multidisciplinary Perspectives on Menopause,* M. Flint, F. Kronenberg, and W. Utian (eds.) (New York, NY: New York Academy of Sciences, 1990), pp. 263-271.

7. Bush, T.L., Cowan, L.D., Barrett-Connor, E., et al., "Estrogen Use and All Cause Mortality: Preliminary Results From the Lipid Research Clinics Program Followup Study," *Journal of the American Medical Association* 249(7):903-906, *1983.*

8. Bush, T.L., Barrett-Connor, E., Cowan, L.D., et al., "Cardiovascular Mortality and Noncontraceptive Use of Estrogen in Women: Results From the Lipid Research Clinics Program Follow-up Study," *Circulation* 75(6):1102-1109, *1987.*

9. Campos, H., "Differences in Low Density Lipoprotein Subfractions and Apolipoproteins in Premenopausal and Postmenopausal Women," *Journal of Clinical Endocrinology and Metabolism 67(1):30-35, 1988.*

10. Campos, H., Wilson, P.W.F., Jimenez, D., et al., "Differences in Apolipoproteins and Low-Density Lipoprotein Subfractions in Postmenopausal Women on and off Estrogen Therapy: Results From the Framingham Offspring Study," *Metabolism 39(10): 1033-1038, 1990.*

11. Cattanach, J., "Oestrogen Deficiency After Tubal Ligation," *Lancet* 1(8433):847-849, 1985.

12. Chakravarti, S., Collins, W.P., and Newton, J.R., "Endocrine Changes and Symptomatology After Oophorectomy in Premenopausal Women," *British Journal of Obstetrics and Gynecology 84(10):769-775, 1977.*

13. Colditz, G.A., Brigham and Women's Hospital, Boston, MA, personal communication, 1991.

14. Colditz, G.A., Willett, W. C., Stampfer, M.J., et al., "Menopause and the Risk of Coronary Heart Disease in Women," New *England Journal of Medicine 316(18)* :1105-1110, *1987.*

15. Colditz, G.A., Stampfer, M.J., Willett, W. C., et al., "Prospective Study of Estrogen Replacement Therapy and Risk of Breast Cancer in Postmenopausal Women," *Journal of the American Medical Association* 264(20):2648-2653, 1990.

16. Egeland, G.M., Kuller, L.H., Matthews, K.A., et al., "Hormone Replacement Therapy and Lipoprotein Changes During Early Menopause," *Obstetrics and Gynecology 76(5, Pt.* 1):776-782,1990.

17. Ernster, V.L., Bush, T.L., Huggins, G.R., et al., "Benefits and Risks of Menopausal Estrogen and/or ProgestinHormone Use," *PreventiveMedicine 17:301-323, 1988.*

18. *Gennari, C.,* Agnusdei, D., Nardi, P., et al., "Estrogen Preserves a Normal Intestinal Responsiveness to l,25-Dihydroxyvitamin D_3 in Oophorectomized Women," *Journal of Clinical and Endocrinologic Metabolism 71(5):1288-1293,1990.*

19. Gordon, T., Kannel, W.B., Hjortland, M.C., et al., "Menopause and Coronary Heart Disease: The Frarningham Study," *Annals of Internal Medicine* 89(2):157-161, *1978.*

20. Henderson, B.E., Paganini-Hill, A., and Ross, R.K., "Decreased Mortality in Users of Estrogen Replacement Therapy," *Archives of Internal Medicine 151:75-78,* 1991.

21. Henderson, M.M., Kushi, L.H., Thompson, D.J., et al., "Feasibility of a Randomized Trial of Low-Fat Diet for the Prevention of Breast Cancer: Dietary Compliance in the Women's Health Trial Vanguard Study," *Preventive Medicine* 19:115-133, *1990.*

22. Hjortland, M.C., NcNamara, P. M., and Kannel, W. B., "Some Atherogenic Concomitants of Menopause," *American Journal of Epidemiology* 103(3): *304-311, 1976.*

23. Hreshchyshyn, M.M., Hopkins, A., Zylstra, S., et al., "Effects of Natural Menopause, Hysterectomy, and Oophorectomy on Lumbar Spine and Femoral Neck Bone Densities," *Obstetrics and Gynecology* 72(4):631-638,1988.

24. Kanders, B., Dempster, D.W., and Lindsay, R., "Interaction of Calcium Nutrition and Physical Activity on Bone Mass in Young Women," *Journal of Bone Mineral Research 3(2); 145-149, 1988.*

25. Kannel, W.B., "Oral Contraceptive Hypertension and Thromboembolism,' *International Journal of Gynecology and Obstetrics* 16:466-472, *1979.*

26. Kaufert, P.L., "Methodological Issues in Menopause Research," *Annals of the New York Academy of Sciences, vol. 592, Multidisciplinary Perspectives on Menopause,* M. Flint, F. Kronenberg, and W. Utian (eds.) (New York, NY: New York Academy of Sciences, 1990), pp. 114-122.

27. Kelsey, J.L., and Hildreth, N. G., *Breast and Gynecologic Cancer Epidemiology* (Boca Raton, FL: CRC Press, 1983).

28. Kemnitz, J.W., Gibber, J.R., Lindsay, K.A., et al., "Effects of Ovarian Hormones on Eating Behaviors, Body Weight, and Glucoregulation in Rhesus Monkeys," *Hormones and Behavior* 23:235-250, *1989.*

29. Khan, S. S., Nessim, S., Gray, R., et al., "Increased Mortality of Women in Coronary Artery Bypass Surgery: Evidence for Referral Bias," *Annals of Internal Medicine* 112(8):561-567,1990.

30. Kirschstein, R.L., "Research on Women's Health," *American Journal of Public Health* 81(3):291-293, *1991.*

31. Kronenberg, F., "Hot Flashes: Epidemiology and Physiology," *Annals of the New York Academy of Sciences,* vol. *592, Multidisciplinary Perspectives on Menopause,* M. Flint, F. Kronenberg, and W. Utian (eds.) (New York, NY: New York Academy of Sciences, 1990), pp. 52-86.

32. **Kuller, L.H.,** Gutai, J.P., **Meilahn,** E., et al., "Relationship of Endogenous Sex Steroid Hormones to Lipids andApoproteins in Postmenopausal Women,' *Atherosclerosis* 10(6):1058-1066, 1990.

33. LaRosa, J., George Washington University Medical Center, Washington, DC, personal communication, September 1991.

34. **Longcope,** C., Herbert, P.N., **McKinlay, S.M.,** et al., "The Relationship of Total and Free Estrogens and Sex Hormone-Binding Globulin With Lipoproteins in Women,' *Journal of Clinical Endocrinology and Metabolism* 71(1):67-72,1990.

35. Manson, J.E., **Colditz, G.A., Stampfer, M.J.,** et al., "A Prospective Study of Obesity and Risk of Coronary Heart Disease in Women," New *England Journal of Medicine* 322(13):882-889, *1990.*

36. Matthews, K.A., **Meilahn,** E., **Kuller, L.H.,** et al., "Menopause and Risk Factors for Coronary Heart Disease," *New EnglandJournal of Medicine* 321(10): *641-646,1989.*

3'7. **McKinlay, S.M.,** New England Research Institute, Watertown, MA, personal communication, October 1991.

38. National Institutes of Health Consensus Conference, "Osteoporosis," *Journal of the American Medical Association* 252:799-802, *1984.*

39. Notelovitz, M., "Carbohydrate Metabolism in Relation to Hormonal Replacement Therapy," *Acta Obstetrics and Gynecology Scandia* 106(Suppl.):51-56, 1982.

40. **Notelovitz,** M., "Exercise and Health Maintenance in Menopausal Women," *Annals of the New York Academy of Sciences,* vol. *592, Multidisciplinary Perspectives on Menopause,* M. Flint, F. **Kronen**berg, and W. Utian (eds.) (New York, NY: New York Academy of Sciences, 1990), pp. 204-220.

41. Notelovitz, M., Women's Medical and Diagnostic Center and the Climacteric Clinic, Gainesville, FL, personal communication, 1991.

42. **Ostlund,** R.E., Staten, M., Kohrt, W.M., et al., "The Ratio of Waist-to-Hip Circumference, Plasma Insulin Level, and Glucose Intolerance as Independent Predictors of the HDL2 Cholesterol Level in Older Adults,'' *New England Journal of Medicine* 322(4):229-234, 1990.

43. Resnick, N.M., and Greenspan, S. L., "'Senile Osteoporosis' Reconsidered," *Journal of the American Medical Association* 261(7):1025-1029, *1989.*

44. Roos, N.P., "Hysterectomies in One Canadian Province: A New Look at Risks," *American Journal of Public Health* 74(1):39-46,1984.

45. Rosenberg, L., ''**Methodologic** Considerations in the Study of Menopause and Its Treatment," contractor report prepared for the Office of Technology Assessment, Nov. 25, 1991.

46. Rothman, K.J., *Modern Epidemiology* (Boston, MA: Little, Brown and Co., 1986).

47. Sherman, S. S., "Research Needs on the Effects of Menopause on the Health of Women," contractor report prepared for the Office of Technology Assessment, Apr. 4, 1991.

48. Shock, N.W., **Gruelich,** R. C., Andres, R.A., et al. (eds.), "Design and Operation of the Baltimore Longitudinal Study of Aging," *Normal Human Aging: The Baltimore Longitudinal Study of Aging,* NIH Pub. 84-2450 (Washington, DC: U.S. Government Printing Office, 1984), pp. 45-57.

49. Siddle, N., **Sarrel,** P., and Whitehead, M., "The Effect of Hysterectomy on the Age at Ovarian Failure: Identification of a Subgroup of Women With Premature Loss of Ovarian Function," *Fertility and Sterility* 47(1):94-100, *1987.*

50. *Smith,* K., **Tremin** Trust, Salt Lake City, UT, personal communication, 1991.

51. **Stampfer, M.J.,** Brigham and Women's Hospital, Boston, MA, personal communication, 1991.

52. **Stampfer, M.J., and Colditz, G.A.,** "Estrogen Replacement Therapy and Coronary Heart Disease: A Quantitative Assessment of the Epidemiologic Evidence," *Preventive Medicine* 20:47-63, *1991.*

53. **Stampfer, M.J., Colditz, G.A.,** and Willett, W. C., "Menopause and Heart Disease: A Review,' *Annals of the New York Academy of Sciences,* vol. *592, Multidisciplinary Perspectives on Menopause,* M. Flint, F. Kronenberg, and W. Utian (eds.) (New York, NY: New York Academy of Sciences, 1990), pp. 193-203.

54. **Stampfer, M.J., Colditz, G.A., Willett,** W. C., et al., "Postmenopausal Estrogen Therapy and Cardiovascular Disease: Ten-Year Followup From the Nurses' Health Study," New *England Journal of Medicine* 325(11):756-762, *1991.*

55. Steering Committee of the Physician's Health Study Research Group, "Final Report on the Aspirin Component of the Ongoing Physician's Health Study," *New England Journal of Medicine* 321:129-135, 1989.

56. Steinberg, K.K., Thacker, S.B., Smith, J., et al., "A Meta-Analysis of the Effect of Estrogen Replacement Therapy on the Risk of Breast Cancer," *Journal of the American Medical Association* 265(15):1985-1990, 1991.

57. Thomas, D. B., Fred Hutchinson Cancer Research Center, Seattle, WA, personal communication, September 1991.

58. U.S. Congress, General Accounting Office, "National Institutes of Health: Problems in Implementing Policy on Women in Study Populations," testimony of Mark V. Nadel before the Subcommittee on Housing and Consumer Interest, Select Committee

on Aging, U.S. House of Representatives, July 24, 1990.

59. U.S. Congress, Office of Technology Assessment, *Policy Issues in the Prevention and Treatment of Osteoporosis* (Washington, DC: U.S. Government Printing Office, forthcoming).

60. U.S. Public Health Service, Department of Health and Human Services, *Action Plan for Women's Health, 1990.*

61. *van* Dam, S., Gillespy, M., Notelovitz, M., et al., "Effect of Exercise on Glucose Metabolism in Postmenopausal Women," *American Journal of Obstetrics and Gynecology* 159(1):82-86,1988.

62. Vandenbroucke, J.P., "Postmenopausal Oestrogen and Cardioprotection,' *Lancet* 337:833-834, 1991.

63. Wade, G.N., and Gray, J.M., "Theoretical Review. Gonadal Effects on Food Intake and Adiposity: A Metabolic Hypothesis," *Physiology and Behavior* 22(3):583-593, *1979.*

64. Wenger, N.K., "Gender, Coronary Artery Disease, and Coronary Bypass Surgery," *Annals of Internal Medicine* 112(8):557-558,1990.

65. Wilson, P.W.F., Garrison, R.J., and Castelli, W.P., "Postmenopausal Estrogen Use, Cigarette Smoking and Cardiovascular Morbidity in Women Over 50: The Framingham Study," *New England Journal of Medicine* 313(17):1038-1043, *1985.*

66. Wing, R.R., Matthews, K.A., Kuller, L.H., et al., "Weight Gain at the Time of Menopause," *Archives of Internal Medicine* 151:97-102,1991.