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# HRT and Genetics: A Role for Polymorphisms?

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*Recent findings suggest a genetic variant in an estrogen-receptor gene may result in a doubling of HDL cholesterol levels in women who take estrogen replacement therapy after menopause. The findings raise interesting possibilities with regard to the potential usefulness of genetic testing, but also raise questions about how increased responsiveness to estrogen may affect other domains of estrogen action.*

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## Introduction

For menopausal women, hormone replacement therapy (HRT)—either estrogen alone or in combination with a progestin—can bring relief from menopausal symptoms such as hot flashes. Estrogen therapy has been demonstrated to lower low-density lipoprotein (LDL) cholesterol plasma levels and raise levels of high-density lipoprotein (HDL) cholesterol.<sup>1,2</sup>

One of the issues in the use of HRT and its effect on cholesterol is determining how an individual woman will respond to the therapy. Estrogen administration has varying effects on HDL cholesterol levels in women. According to studies of families, variability in HDL cholesterol levels may have a genetic foundation.<sup>3</sup> Thus, an intriguing area to be explored is the link between hormone replacement, HDL cholesterol levels and genetics.

It has already been established that there are genetic variants in many of the steroid hormone receptor genes, including the vitamin D, glucocorticoid receptor and androgen-receptor genes; however, less is known about the estrogen-receptor genes. If a common polymorphism were found in the estrogen-receptor genes (alpha or beta) that altered individual responses to estrogen, it could

have important implications regarding the safety and efficacy of HRT, and add to our understanding of estrogen action.

To address this issue, we conducted a study of the association between sequence variants in the estrogen-receptor-alpha (ER- $\alpha$ ) gene and the response of the HDL cholesterol level to HRT.<sup>4</sup> The study group was taken from the Estrogen Replacement and Atherosclerosis (ERA) trial.<sup>5</sup>

The ERA trial was an attempt to ascertain the role of estrogen and progestin in the treatment of heart disease. A total of 309 women with angiographically verified coronary artery disease were randomized to one of three regimens: 0.625 mg of conjugated estrogen per day, 0.625 mg of conjugated estrogen plus 2.5 mg of medroxyprogesterone acetate per day, or placebo. The women were followed for just over three years, and coronary angiograms were analyzed by quantitative coronary angiography.

Both hormone regimens resulted in significant reductions in LDL cholesterol and increases in HDL cholesterol. However, neither regimen slowed the progression of atherosclerosis. The rates of clinical cardiovascular events were similar among the treatment groups. We concluded that the hormone treatment did

not affect the progression of coronary atherosclerosis in women with established disease, and that these women should not use estrogen replacement for the purpose of achieving cardiovascular benefit.

## Estrogen Receptor Polymorphisms

For the estrogen gene polymorphism study, we performed DNA isolation and genotyping of women from the ERA trial. We were looking for the presence or absence of a variant in the ER- $\alpha$  gene and its effect on HDL.

We looked at a total of 10 different variants that were scattered throughout the ER- $\alpha$  gene. It is estimated that there are variants that occur almost every 1,000 base pairs in DNA. The estrogen-receptor gene is nearly a quarter of a million base pairs in size, and many more of its variants have not yet been described.

## Results

Women who had one specific variant in their ER- $\alpha$  gene (the ER- $\alpha$  IVS1-401 C/C genotype) had double the increase in HDL cholesterol levels when treated with estrogen or estrogen plus progestin: 13.1 mg/dl for women with the variant gene, vs 6 mg/dl for those without the gene.

Another remarkable aspect about find-

ing this variant is its size. In the genetic blueprint of the human genome, there are roughly 3 billion letters. This estrogen-receptor variant involves the change of a *single* letter. A change of one single base pair out of 3 billion was capable of identifying women who respond dramatically to estrogen with respect to HDL, and possibly other things.

There are three important points that can be made about this finding:

- **This genetic variant is common.**

Many genetic studies have looked at gene polymorphisms that occur in 0.5 to 1% of the population, and so their clinical relevance is not great. In this case, the genetic variant of interest occurred in 20% of the women studied. In other words, 1 in 5 women are likely to have this genetic predisposition to higher HDL cholesterol when taking estrogen.

- **The magnitude of the increase in HDL in these women was dramatic.** The increase was two or three times greater than what is typically seen with other drugs that are taken specifically for the purpose of raising HDL.

- **Women with this genetic variant may be more sensitive to estrogen in other domains of estrogen action.** For instance, this could include favorable reactions on bone. On the other hand, it might also include some adverse effects, such as an increased risk for venous thrombosis or breast cancer. We did find that estrogen-treated women with this ER- $\alpha$  variant had greater reductions in the adhesion molecule E-selectin and no increase in the inflammation marker C-reactive protein, both of which suggest favorable effects on the cardiovascular system.<sup>6</sup>

There was no prior hypothesis that this particular gene polymorphism would influence HDL in this manner. However, because it is known that gene variants show altered functions, it was not unexpected to find variants in the estrogen-receptor gene that would alter the function of estrogen.

### The Implications

The three previous points lead us to be-

lieve that these findings might be of true clinical significance. These are very promising data, in part because of the impressive increase in HDL that we observed. However, we do not know yet whether these findings will translate into dramatic reductions in risk for heart disease. The answer to that question awaits further studies. The increase in HDL levels with HRT use in patients with this variant may not correlate with cardiovascular event benefit.

A potential problem is that researchers have been unable to document that HRT lowers the risk of heart disease for women who already have heart disease. We have cited data from the ERA trial. If the progression rate in the active treatment arms were in a beneficial direction, but the confidence intervals still included the rates in the placebo group, then we could speculate about the presence of a more subtle effect that might have been detected with another technique. However, two points argue against that interpretation: (1) The point estimates were virtually identical, and (2) there was no evidence of a greater benefit with a longer treatment period. It should be noted that angiography has been a remarkably useful technique to predict efficacy of anti-atherosclerotic interventions.

In another trial, the Heart and Estrogen/progestin Replacement Study (HERS), investigators set out to determine if estrogen plus progestin therapy alters the risk for coronary heart disease events in postmenopausal women with established coronary disease.<sup>7</sup>

Participants were given either 0.625 mg of conjugated equine estrogens plus 2.5 mg of medroxyprogesterone acetate, or a placebo, and followed for an average of 4.1 years. HRT use did not reduce the overall rate of coronary heart disease events. Overall, there were no significant differences between groups in the primary outcome or in any of the secondary cardiovascular outcomes; 172 women in the hormone group and 176 women in the placebo group had myocardial infarction or coronary heart disease death. The women in the hormone group did have a

net 11% lower LDL cholesterol level and 10% higher HDL cholesterol level. Based on these findings, the investigators did not recommend HRT for secondary prevention of coronary heart disease.

### Estrogen and Heart Disease

The fact that the studies to date have largely been negative does not rule out the possibility that HRT still may have an important clinical effect. It is possible that in this subgroup of women, because of the enhanced effect on HDL, there is some benefit. We might have been unable to see it because it might have been diluted among the other women in the cohort who did not have the gene polymorphism.

A great deal of work remains before we can establish the practical outcomes of these findings. We need to look specifically in women with and without this gene polymorphism to make a judgment about their clinical outcomes. We also need to examine the effects of this gene polymorphism on the broad range of health outcomes that are influenced by estrogen before we can be certain what role screening for this polymorphism might play in clinical practice.

The next critical step is to confirm or refute these findings in other studies that will enable us to look at clinical events. One approach would be to genotype the participants in the HERS trial. We must also conduct prospective clinical trials of women who are randomized to receive HRT or placebo, and then look among those receiving hormone therapy to see if the outcome was different, depending on genotype.

### The Future

For primary-care physicians, the message appears to be that we may soon be able to use simple genetic testing to differentiate individual responses to HRT. Ultimately, we may be able to identify women who are more likely to benefit from estrogen. Tests for gene polymorphisms like the one we have described, and possibly others like it, may become commonplace in the practice of medicine.

Other gene polymorphisms have been

described that modify the effects of many of the other commonly used medicines in clinical practice, including the effects of statins and medications used to treat blood pressure and asthma. Our study illustrates the kind of testing that we expect will become a common part of the practice of medicine in the near future.

Within the next 5 years, we expect to see recommendations about the use of genetic testing to improve selection of the type or dose of many different drugs commonly used in clinical practice. Ultimately, in the case of HRT, the availability of testing for polymorphisms may help clinicians make smarter decisions regarding to whom they prescribe estrogen and how much they prescribe. ■

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## Hot Flash Etiology: New Directions for Research

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and on hormonal status and thermoregulation, but women going through menopause are experiencing both. Plans are under way to conduct research in this area in the near future. Additional research also is needed in the area of hot flashes and their effect on sleep. To date, only two studies have been conducted in this area, both of which showed that half of the hot flashes documented were not, in fact, accompanied by awakenings.<sup>18,19</sup>

Ultimately, a better understanding of the etiology of hot flashes will undoubtedly affect the way in which new pharmaceuticals are developed. For example, drugs like clonidine that reduce sympathetic activation but do not cause sedation or hypotension would be of great value. In the meantime, ongoing research is bringing us closer to an understanding of estrogen's mechanism of action in the treatment of hot flashes, and is leading us in new and exciting directions. ■

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