Measuring Urinary Estrogen Metabolites for Cancer Risk Assessment

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Testing for the urinary levels of 2-hydroxyestrone and 16 alpha–hydroxyestrone provides valuable insight regarding disease risk and can be treated with nutritional interventions[1-3]. The ratio of 2-hydroxyestrone (2OHE1) to 16 alpha-hydroxyestrone (16OHE1), or the Estrogen Metabolite Index (EMI), should be greater than 2.0 and values in the upper normal range are advisable. Any woman using HRT who has a low EMI should be monitored closely for improvements in the urinary metabolites. Women who have a low EMI may be treated with Indole-3-carbinol (I3C) and diindolylmethane (DIM) supplements, sulfur containing supplements, as well as with ground flaxseed and soy.

Why test for the 2:16 Ratio?

The major metabolites of estradiol and estrone are those hydroxylated at either the C-2 or the C-16 positions. 2-hydroxyestrone (2OHE1) is essentially devoid of peripheral biological activity, as shown in studies on uterine weight, gonadotrophin secretion, and cell proliferation[1]. 2OHE1 has even been found to exert a modest anti-estrogenic effect,[3,4] and has been called "the good estrogen"[5]. The 16 alpha-hydroxylated metabolites, 16 alpha-hydroxyestrone and estriol (E3), are estrogen agonists[2]. Women with breast and endometrial cancers have marked elevation of 16 alpha-hydroxylation, which is a significant risk factor for such estrogen-dependent tumors[6]. Tumors in other estrogen-sensitive tissues are also promoted by 16OHE1. For example, as the ratio of 2OHE1 to 16OHE1 decreases, the severity of recurrent respiratory papillomatosis also increases[7].

The ratio of 2-hydroxyestrone to 16 alpha-hydroxyestrone is not only a risk factor of breast cancer but also other conditions of inappropriate estrogen activity. 16 alpha-hydroxyestrone stimulates cell mitosis and proliferation, sometimes contraindicated in certain diseases. 16 alpha-hydroxyestrone has been found to be elevated in those at risk for breast cancer, as well as other conditions associated with hyperimmune activity such as systemic lupus erythematosis and rheumatoid arthritis. In these populations 16 alpha-hydroxyestrone was 10 times higher than the control population[8]. Estrogen metabolism should therefore be evaluated when treating patients with autoimmune conditions.

Cancers that react favorably to a higher 2:16 ratio

- ER + breast cancer
- ER – breast cancer
- Prostate cancer
- Cervical cancer
- Ovarian cancer
- Laryngeal cancer

Because there is an optimal level of both 2- and 16-hydroxyestrone, a ratio comparing the two levels can be very helpful. A 2:16 ratio is highly correlated with breast cancer. In 10,786 women, followed for 5.5 years, higher 2:16 ratios correlated with lower risk of breast cancer[13]. Altering the ratio with I3C showed promising therapeutic effect not only for estrogen receptor positive cancers but also for estrogen receptor negative cancers[11]. Research regarding the 2:16 ratio was also of clinical value for other cancers as well. In a group of 8 cervical cancer patients, 4 out of 8 had complete remission as their 2:16 ratio improved in a dose-dependent fashion. The greatest response was seen in the group taking 200 mg/day of I3C[14]. Because metabolites are being considered, the preferred method for measuring 2:16 ratio is in the urine. 2:16 ratio does not fluctuate with timing of the menstrual cycle or with menopausal status[15].

Improving the 2:16 Ratio

Unlike certain risk factors for cancer such as genetics, a 2:16 ratio is highly treatable. Indole-3 carbinole Measuring Urinary Estrogen Metabolites significantly improves a 2:16 ratio[9, 10]. A decreased risk of metastasis was found with an improved ratio[11]. Lifestyle factors such as exercise and a high protein diet were found to improve

ratios[12]. Even though a large body of research exists demonstrating safety with I3C, more recent studies may begin to favor DIM, a derivative of I3C because it appears not to increase 4OHE1.

The increased risk from estrogen and estrogen metabolites has led to a search for compounds that produce estrogen-like effects safely and those that decrease the production of 16OHE1. Women who have a low EMI may be treated with I3C and DIM supplements, sulfur containing supplements, as well as with ground flaxseed and soy. Soy isoflavones (e.g., daidzein and genistein) are natural compounds increased by eating a diet rich in soy products[16]. Soy isoflavones have little or no affect on induction of tumors, and they have many estrogen-like properties even though they operate through different receptor sites on target tissues[17-19]. Dietary intake of soy products[20] and flax[21] have been shown to favorably modulate the rates of 2- vs. 16-hydroxyestrone production.

Indole-3-carbinol (I3C) and diindolylmethane (DIM) also help to modulate estrogen metabolism. Estrogens are metabolized by cytochrome P-450 (CYP450) enzymes that are inducible by compounds found in vegetables such as cabbage, Brussels sprouts, and broccoli,[10] from the Brassica plant family. Two phytochemicals contained in these foods, I3C and DIM, have been identified as active inducers of certain P450-isozymes, particularly CYP1A1[22].

The reaction catalyzed by P4501A1 produces 2-hydroxylation of estradiol. Induction of P4501A1 causes a competitive down-regulation of P4501B1, which is the enzyme responsible for producing 16OHE1. Therefore, if P4501A1 enzyme is not sufficiently active, CYP1B1 activity will increase the estrogen agonist metabolite 16OHE production and drive down the EMI.

However, there is some recent evidence that suggests that I3C may increase 4-hydroxylation of estrone and estradiol, whereas DIM may not. 4-hydroxylation has been demonstrated to promote breast and prostate cancer tissue via estrogen receptor stimulation as well as DNA damage. Additionally, 4OHE is elevated in breast cancer patients[23]. However, 4OHE1 is an extremely minor metabolite, comprising less than 1% of estrogen metabolism, thus whether this metabolite or the DNA adducts formed by its activity are important markers in cancer risk assessment still remains to be elucidated.

Other constituents in the cruciferous family are also speculated to aid in estrogen metabolism. Glutathione S transferase also appears to be upregulated by the sulfur constituents in cruciferous vegetables. Brassica vegetables also improve glucuronidation aiding with elimination of estrogen metabolites. These compounds that aid estrogen metabolism were also found to decrease DNA damage, quantifiable by reduction in 8-OH 2-deoxyguanosine, an oxidative marker of DNA damage. Flaxseed supplementation at 10 g/d significantly increases the urinary 2/16 hydroxy-estrone ratio, suggesting a breast cancer chemo-protective effect[24].

The Estronex™ test, measuring estrogen metabolism, is useful for clinicians seeking to prevent cancer occurrence or reoccurrence as well as other diseases. It is recommended to encourage the production of 2OHE1 with its anti-estrogenic effect,[3,4] rather than 16 alpha-hydroxyestrone with its estrogen agonistic effect[2]. I3C and DIM supplements, sulfur containing supplements, as well as ground flaxseed and soy have been used to modulate the 2:16 ratio, but some speculate that I3C has adverse consequences.

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