

Sexual Tension in the Airways The Puzzling Duality of Estrogen in Asthma

Yes, reader, as you probably suspected, the double-entendre in our title is merely a marketing ploy, aimed to draw your attention to this issue's intriguing article about estrogen effects on airway responsiveness by Matsubara and coworkers (1). While there will be no further mention of sexual tension or other topics in reproductive physiology, here is why this article merits your interest.

There are important sex differences in the natural history of asthma, repeatedly identified clinically and epidemiologically. The core observation is that in pre-pubescent children, asthma is more common in males, but after puberty, asthma is more common in females (2, 3). The magnitude of this sex difference is perhaps best described as modest (post-pubertal male versus female odds ratio 0.8 [3], or 44.4% of males with current asthma were diagnosed at 18 yr or older versus 63.9% for females [4]). However, given the frightfully large number of individuals with asthma, these percentage shifts have considerable impact. In addition to prevalence, asthma morbidity is greater in females than in males, as measured by risk of hospitalization (5, 6). A role for sex hormones in modulating asthma can be deduced from these sex differences, and more directly from increased asthma exacerbations during menses (7, 8), effects of hormone-replacement therapy on asthma diagnosis (9, 10), and airway function in post-menopausal women (11, 12).

It is fair to say that the story gets more complicated once we turn to the experimental data. Some studies easily reproduce the general trend of the human epidemiologic data, especially those focusing on immune and inflammatory measures of allergy. Using standard protocols of systemic sensitization with ovalbumin (OVA) and airway challenge, many investigators find that female mice show greater allergic lung disease as compared with males, with increased OVA-specific immunoglobulin IgG1 and IgE, increased Th2 cytokines, and increased peripheral blood eosinophilia (13–16). Surgical manipulation provides specific data implicating estrogen as the basis of this sex difference. Ovariectomized (OVX) rats developed less airway inflammation than sham-operated controls, indicating that female sex hormones play a pro-inflammatory role. Replacement of estradiol re-established airway inflammation in the OVX rats, and treatment of intact rats with tamoxifen (estrogen receptor antagonist) attenuated development of airway inflammation (17). The same group also showed that bone marrow cells of OVX rats treated with estradiol released more IL-4 and less IL-10 compared with OVX rats not so treated. Bronchoalveolar lavage (BAL) cells of the estradiol-treated groups also had increased release of IL-1 β and TNF- α (18).

More detailed analysis has revealed an interesting dichotomy: while estrogen enhances the allergic inflammation in asthma, it also acts to down-regulate airway hyperresponsiveness (AHR). Males of the C57BL/6 and BALB/c strains are more sensitive than the females to inhaled methacholine, showing greater changes in total respiratory system resistance, elastance, and other mechanical parameters. Estrogen receptor α -deficient mice have increased airway responsiveness to methacholine and

serotonin. When made allergic, these mice have increased airway responsiveness (19), indicating that estrogen acts to decrease AHR. There are a number of proposed mechanisms for this effect, including decreased acetylcholine-induced airway reactivity by increased epithelial acetylcholinesterase activity (20) and relaxation of tracheal smooth muscle cells by opening of BKCa (large conductance, calcium-activated potassium channels) through activation of the nitric oxide–cGMP–protein kinase G pathway (21).

In this issue, Matsubara and colleagues (1) use a murine model to further characterize how estrogens can suppress AHR, and also identify a possible novel mechanism. Using a “lung exposure only” protocol (10 d nebulized OVA, without systemic sensitization or adjuvant), Matsubara and coworkers (1) demonstrate that estrogen is protective against *in vivo* AHR. Only male mice developed *in vivo* AHR after their exposure protocol. Estradiol suppressed the AHR in a dose-dependent manner, and this suppression was reversed with an estrogen receptor antagonist. They also showed that OVX mice, but not sham-operated females, developed AHR, which was reversed with estradiol, and that estrogen receptor antagonists in non-OVX females induced AHR. All of these data provide a robust demonstration that estrogens are protective in this experimental model. This model does not generate a significant cellular inflammatory response and there were no differences in BAL or cytokine parameters between groups.

This study also addressed in more detail the mechanism by which estrogen may be acting. Even when given to males as little as 1 hour before assay of airway responsiveness, estrogen suppressed AHR. Similarly, when an estrogen receptor antagonist was given to females 1 hour before assay, AHR was enhanced. These data, along with the lack of potentially confounding influence of airway inflammation in this unusual variant of mouse asthma models, allow a focus on direct effects of estrogens on airway contractility. Matsubara and colleagues further show that an NK-1 receptor antagonist (Sendide) had similar effects, with inhibition of AHR, supporting the study's conclusion that, at least in this model, estrogens suppress AHR via an NK-1–dependent pathway.

The mechanistic link to neurokinin biology is supported by the precedent of estrogen regulation of smooth muscle contractility in the uterus via effects on neurokinin receptor desensitization (22). We wonder if future studies should not also look for lessons from endothelial biology. Here, estrogen also modulates smooth muscle contractility in a relatively rapid, nongenomic manner by enhancing activity of endothelial nitric oxide synthase, a process that is vasodilatory and atheroprotective in females (23). The expression of endothelial nitric oxide synthase in airway epithelium (24) may allow similar effects on bronchial smooth muscle, and it is noteworthy that eNOS-deficient mice have been reported to manifest AHR (25).

Some limitations of the data specific to this article must be considered, but the reader should be aware of two complexities in this field, which we can only briefly mention. First, the literature is bedeviled by numerous observations that estrogen effects can vary significantly depending on dosage and timing of administration (26, 27). Second, and more important biologically, there are ample data that other hormones, especially androgens, modulate allergic

airway disease and may protect against asthma (14). Turning to the model used by Matsubara and colleagues, it is noteworthy that it does not use sensitization nor adjuvant, and provides a model of AHR with minimal allergic inflammation. While this is convenient and experimentally clever for isolating AHR as the experimental variable, it does make this murine system quite removed from the human disease it seeks to model. The key finding that estrogens act to down-regulate AHR is difficult to reconcile with the available human data, which indicate that overall estrogens are pro-asthmatic (9, 10, 12). It should be noted, however, that some human data do show protective effects of estrogen (28–30). Moreover, the detailed comparison of sex/estrogen effects on AHR versus allergic inflammation, which is possible in animal models, would be difficult to achieve in human studies. Hence, how well these findings extrapolate to human airways remains an open question. Nevertheless, the mechanistic insights offered will help guide further experimentation and the search for potentially novel therapeutics for individuals with asthma of both sexes.

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