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Dockets Management Branch (HFA-305)
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To whom it may concern,

We are writing in reference to Docket # 98P-0683; "Food Labeling: Health Claims; Soy Protein and Coronary Heart Disease". We oppose this health claim because there is abundant evidence that some of the isoflavones found in soy, including genistein and equol, a metabolite of daidzen, demonstrate toxicity in estrogen sensitive tissues and in the thyroid. This is true for a number of species, including humans. Additionally, the adverse effects in humans occur in several tissues and, apparently, by several distinct mechanisms.

Genistein is clearly estrogenic; it possesses the chemical structural features necessary for estrogenic activity (; Sheehan and Medlock, 1995; Tong, et al, 1997; Miksicek, 1998) and induces estrogenic responses in developing and adult animals and in adult humans. In rodents, equol is estrogenic and acts as an estrogenic endocrine disruptor during development (Medlock, et al, 1995a,b). Faber and Hughes (1993) showed alterations in LH regulation following developmental treatment with genistein. Thus, during pregnancy in humans, isoflavones *per se* could be a risk factor for abnormal brain and reproductive tract development. Furthermore, pregnant Rhesus monkeys fed genistein had serum estradiol levels 50-100% higher than the controls in three different areas of the maternal circulation (Harrison, et al, 1998). Given that the Rhesus monkey is the best experimental model for humans, and that a women's own estrogens are a very significant risk factor for breast cancer, it is unreasonable to approve the health claim until complete safety studies of soy protein are conducted. Of equally grave concern is the finding that the fetuses of genistein fed monkeys had a 70% higher serum estradiol level than did the

controls (Harrison, et al, 1998). Development is recognized as the most sensitive life stage for estrogen toxicity because of the indisputable evidence of a very wide variety of frank malformations and serious functional deficits in experimental animals and humans. In the human population, DES exposure stands as a prime example of adverse estrogenic effects during development. About 50% of the female offspring and a smaller fraction of male offspring displayed one or more malformations in the reproductive tract, as well as a lower prevalence (about 1 in a thousand) of malignancies. In adults, genistein could be a risk factor for a number of estrogen-associated diseases.

Even without the evidence of elevated serum estradiol levels in Rhesus fetuses, potency and dose differences between DES and the soy isoflavones do not provide any assurance that the soy protein isoflavones *per se* will be without adverse effects. First, calculations, based on the literature, show that doses of soy protein isoflavones used in clinical trials which demonstrated estrogenic effects were as potent as low but active doses of DES in Rhesus monkeys (Sheehan, unpublished data). Second, we have recently shown that estradiol shows no threshold in an extremely large dose-response experiment (Sheehan, et al, 1999), and we subsequently have found 31 dose-response curves for hormone-mimicking chemicals that also fail to show a threshold (Sheehan, 1998a). Our conclusions are that no dose is without risk; the extent of risk is simply a function of dose. These two features support and extend the conclusion that it is inappropriate to allow health claims for soy protein isolate.

Additionally, isoflavones are inhibitors of the thyroid peroxidase which makes T3 and T4. Inhibition can be expected to generate thyroid abnormalities, including goiter and autoimmune thyroiditis. There exists a significant body of animal data that demonstrates goitrogenic and even carcinogenic effects of soy products (cf., Kimura et al., 1976). Moreover, there are significant reports of goitrogenic effects from soy consumption in human infants (cf., Van Wyk et al., 1959; Hydovitz, 1960; Shepard et al., 1960; Pinchera et al., 1965; Chorazy et al., 1995) and adults (McCarrison, 1933; Ishizuki, et al., 1991). Recently, we have identified genistein and daidzein as the goitrogenic isoflavonoid components of soy and defined the mechanisms for inhibition of thyroid peroxidase (TPO)-catalyzed thyroid hormone synthesis in vitro (Divi et al., 1997; Divi et al., 1996). The observed suicide inactivation of TPO by isoflavones, through covalent binding to TPO, raises the possibility of neoantigen formation and because anti-TPO is the principal autoantibody present in autoimmune thyroid disease. This hypothetical mechanism is consistent with the reports of Fort et al. (1986, 1990) of a doubling of risk for autoimmune thyroiditis in children who had received soy formulas as infants compared to infants receiving other forms of milk.

The serum levels of isoflavones in infants receiving soy formula that are about five times higher than in women receiving soy supplements who show menstrual cycle disturbances, including an increased estradiol level in the follicular phase (Setchell, et al, 1997). Assuming a dose-dependent risk, it is unreasonable to assert that the infant findings are irrelevant to adults who may consume smaller amounts of isoflavones.

Additionally, while there is an unambiguous biological effect on menstrual cycle length (Cassidy, et al, 1994), it is unclear whether the soy effects are beneficial or adverse. Furthermore, we need to be concerned about transplacental passage of isoflavones as the DES case has shown us that estrogens can pass the placenta. No such studies have been conducted with genistein in humans or primates. As all estrogens which have been studied carefully in human populations are two-edged swords in humans (Sheehan and Medlock, 1995; Sheehan, 1997), with both beneficial and adverse effects resulting from the administration of the same estrogen, it is likely that the same characteristic is shared by the isoflavones. The animal data is also consistent with adverse effects in humans.

Finally, initial data from a robust (7,000 men) long-term (30+ years) prospective epidemiological study in Hawaii showed that Alzheimer's disease prevalence in Hawaiian men was similar to European-ancestry Americans and to Japanese (White, et al, 1996a). In contrast, vascular dementia prevalence is similar in Hawaii and Japan and both are higher than in European-ancestry Americans. This suggests that common ancestry or environmental factors in Japan and Hawaii are responsible for the higher prevalence of vascular dementia in these locations. Subsequently, this same group showed a significant dose-dependent risk (up to 2.4 fold) for development of vascular dementia and brain atrophy from consumption of tofu, a soy product rich in isoflavones (White, et al, 1996b). This finding is consistent with the environmental causation suggested from the earlier analysis, and provides evidence that soy (tofu) phytoestrogens causes vascular dementia. Given that estrogens are important for maintenance of brain function in women; that the male brain contains aromatase, the enzyme that converts testosterone to estradiol; and that isoflavones inhibit this enzymatic activity (Irvine, 1998), there is a mechanistic basis for the human findings. Given the great difficulty in discerning the relationship between exposures and long latency adverse effects in the human population (Sheehan, 1998b), and the potential mechanistic explanation for the epidemiological findings, this is an important study. It is one of the more robust, well-designed prospective epidemiological studies generally available. We rarely have such power in human studies, as well as a potential mechanism, and thus the results should be interpreted in this context.

Does the Asian experience provide us with reassurance that isoflavones are safe? A review of several examples lead to the conclusion "Given the parallels with herbal medicines with respect to attitudes, monitoring deficiencies, and the general difficulty of detecting toxicities with long latencies, I am unconvinced that the long history of apparent safe use of soy products can provide confidence that they are indeed without risk." (Sheehan, 1998b).

It should also be noted that the claim on p. 62978 that soy protein foods are GRAS is in conflict with the recent return by CFSAN to Archer Daniels Midland of a petition for GRAS status for soy protein because of deficiencies in reporting adverse effects in the petition. Thus GRAS status has not been granted. Linda Kahl (202-418-3101) can provide you with details. It would seem appropriate for FDA to speak with a single voice

regarding soy protein isolate.

Taken together, the findings presented here are self-consistent and demonstrate that genistein and other isoflavones can have adverse effects in a variety of species, including humans. Animal studies are the front line in evaluating toxicity, as they predict, with good accuracy, adverse effects in humans. For the isoflavones, we additionally have evidence of two types of adverse effects in humans, despite the very few studies that have addressed this subject. While isoflavones may have beneficial effects at some ages or circumstances, this cannot be assumed to be true at all ages. Isoflavones are like other estrogens in that they are two-edged swords, conferring both benefits and risk (Sheehan and Medlock, 1995; Sheehan, 1997). The health labeling of soy protein isolate for foods needs to considered just as would the addition of any estrogen or goitrogen to foods, which are bad ideas.

Estrogenic and goitrogenic drugs are regulated by FDA, and are taken under a physician's care. Patients are informed of risks, and are monitored by their physicians for evidence of toxicity. There are no similar safeguards in place for foods, so the public will be put at potential risk from soy isoflavones in soy protein isolate without adequate warning and information.

Finally, NCTR is currently conducting a long-term multigeneration study of genistein administered in feed to rats. The analysis of the dose range-finding studies are near-complete or complete now. As preliminary data, which is still confidential, may be relevant to your decision, I suggest you contact Dr. Barry Delclos at the address on the letterhead, call him at 870-543-7372, or email him at <bdelclos@nctr.fda.gov>.

Sincerely,

Daniel M. Sheehan

Daniel R. Doerge

Enclosures

cc: Dr. Bernard Schwetz, Director, NCTR
Dr. Barry Delclos

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