



AMERICAN
BOTANICAL
COUNCIL

Post Office Box 144345
Austin, Texas 78714-4345
Phone 512/926-4900
Fax 512/926-2345
Email: abc@herbalgram.org
www.herbalgram.org

Mark Blumenthal
Editor

Wayne Silverman, PhD
Underwriting Coordinator

Leela Devi, MSN, RN
Mariann Garner-Wizard
Betsy Levy
Heather Oliff, PhD
Risa Schulman, PhD
Densie Webb, PhD
Summary Writer

Karen Robin
Susan McFarland
Coordinators

Dawnelle Malone
Research Assistant

The American Botanical Council provides this summary and the enclosed article as an educational service. By providing this article, ABC does not warrant that the data is accurate and correct, nor does distribution of the enclosed article constitute any endorsement of the information contained or of the views of the authors.

ABC does not authorize the copying or use of the original articles. Reproduction of the summaries is allowed on a limited basis for students, colleagues, employees and/or customers. Other uses and distribution require prior approval.

HERBCLIPTM

FILE: · Red Clover (*Trifolium pratense*)
· Isoflavones

DATE: March 15, 2000

HC 080590

RE: Benefits of Red Clover Extract

Kelly, G., A. Husband, and M. Waring. *Standardized Red Clover Extract Clinical Monograph*. Natural Products Research Consultants, Inc., Seattle, WA 98104. 1998.

Red Clover (*Trifolium pratense*), also known as cow clover, meadow clover, purple clover, beebread and trefoil, is a native of Europe, Asia and Africa, but is grown around the world as a soil-improving crop and as fodder. It has also been used medicinally by Oriental, European and American cultures. This monograph covers the historical use of red clover and the pharmacological, clinical and safety aspects of an extract standardized to its isoflavonoids.

Historical Use

Both the leaves and the flowers of red clover have been used medicinally. An infusion of the flowers has been used by the Chinese as an expectorant and by the Russians for bronchial asthma. The whole plant was used by Native Americans for sore eyes and burns and as a food, while the Europeans used it for liver and digestive ailments. It has also been used to treat edema, psoriasis and eczema.

Composition

There are four notable types of compounds in fresh red clover leaves and flowers: 1) 11 flavonoid glycosides, including daidzein, genistein, formononetin, biochanin, and calycosin (in levels up to 2% of the dry weight); 2) coumestans; 3) volatile oil, consisting of over 40 compounds including methyl salicylate, benzyl alcohol and eugenol; 4) L-dopa caffeic acid conjugates. Other components include: moisture (81%), ash (2%), protein (4%), fiber (2.6%), fat (0.7%), polysaccharides, resins, fatty acids, hydrocarbons, alcohols, chlorophylls, minerals and vitamins.

Isoflavones—Biological Effects

Of these compounds, the isoflavonoids (daidzein, genistein, formononetin and biochanin) have received a great deal of attention for their numerous *in vivo* and *in vitro* effects, listed below.

Estrogenicity: These compounds exert a weak estrogenic effect due to their ability to bind estrogen receptors.

Steroidogenesis: They modulate several enzymes responsible for regulation of sex hormones; inhibit oxidation of steroid hormones; induce production of sex-hormone binding globulin (SHBG).

Antioxidation: They act as potent antioxidants and promote activity of anti-oxidant enzymes.

Calcium metabolism and bone turnover: They promote storage of calcium; maintain bone density.

Cell growth and differentiation: They inhibit a range of enzymes involved in cell growth, including cancer cells.

Anticancer activity: They inhibit growth of numerous prominent cancers *in vitro* and breast cancer in rats; inhibit angiogenesis (proliferation of blood vessels) and cancer cell adhesion.

Cardiovascular effects: Among a wide range of beneficial effects, the isoflavonoids inhibit platelet aggregation, vasodilation and nitric oxide production.

A standardized red clover isoflavonoid extract (SRCE) tablet contains 40 mg of the four isoflavonoids in their aglucone form in 200-225 mg of dried, aqueous-alcohol extract. This extract is made from three cultivars of *T. pratense* selected for their high content and specific proportion of isoflavones. The approximate ratio is 20:12:1:1 biochanin:formononetin:daidzein:genestein.

Pharmacokinetic data

Pharmacokinetic data are limited to that from one study that at the time of the article's publication was not yet published. In 16 men and women challenged with SRCE, it was found that all four isoflavones were present in the plasma, the kinetics of their plasma levels were similar, their concentrations peaked at 5 hours, and their half lives varied between 9 and 12 hours. In the plasma they are conjugated as glucuronides and sulfonates. Since this article, other data have been published.

Metabolic Data

Both animal and human studies indicate that all four isoflavones are modified in the body, the major mechanism being fermentation by gut flora, though the liver may be involved as well. The main structural change is the demethylation of formononetin to daidzein and biochanin to genistein. In humans, there are much higher levels of demethylated than methylated isoflavones in the blood. Daidzein and genistein are additionally broken down into several metabolites, including equol, O-desmethylangolensin, dihydrodaidzein and dihydrogenestein. A urinary profile from a person who had consumed one tablet of SRCE 24 hours earlier is shown.

Clinical Effects

The clinical effects of SRCE in perimenopausal women suffering acute menopausal symptoms have been studied in two unpublished, double-blind, placebo-controlled trials. The first trial, conducted at the Royal Hospital for Women in Sydney, Australia, compared placebo with both one SRCE tablet per day and two SRCE tablets twice a day for three months (total n=36). The second trial, conducted at the Royal North Shore Hospital, Sydney, Australia, compared placebo with one SRCE tablet per day (total n=86) for three months and then crossed over for three months. The outcome measure was weekly self-reporting of incidence and severity of hot flashes. Blood and urine were collected at the beginning and end of the study. In both trials, there was no significant difference in the incidence or severity of hot flashes for any of the treatment groups; both decreased 32-45%. There was a strong correlation between urinary isoflavone concentration and incidence of hot flashes. In the first trial, there was a significant increase in serum HDL cholesterol with one SRCE tablet, but not with four tablets.

Dosage

The recommended dosage of SRCE is two tablets per day, as that dosage delivers the amount of isoflavones equivalent to that provided by a Japanese or vegetarian diet high in legumes (about 30-100 mg per day).

Safety

No mutagenic changes were detected in Ames mutagenicity tests with *Salmonella typhimurium*. Tests assessing hematological abnormalities in bone marrow cells of mice were likewise negative. The acute oral toxicity test, in which one dose of 3,000 mg/kg SRCE was administered to rats, found no clinical abnormalities during a 14 day observation period. When rats were given the same dose daily for 28 days, no adverse effects were observed, and no organ abnormalities were found at autopsy. Data for humans was drawn from the two unpublished clinical studies discussed above. No adverse reactions, uterine bleeding, endometrial thickening or changes in hematological or serum parameters were noted.

Contraindications and Interactions

There have been no studies to show whether isoflavones interact with other steroid therapeutics. However, since isoflavones exert their effects through competitive inhibition of estrogens, caution should be used in supplementing the diet if other steroids (estrogens, progestins or androgens) are being administered.

Precautions

A dosage of one to two tablets should not be exceeded by lactating women, as isoflavones are secreted in the breast milk and should be in the range of that consumed in the diet. The safety of isoflavones for pregnant women has not been established. Pregnant rats given high doses had offspring with delayed onset of puberty and decreased birthweights and anogenital distance, but there was no effect on parturition, rate of stillbirth or pituitary responsiveness to GnRH (gonadotropin releasing hormone). Isoflavones have never been implicated in fetal abnormalities in communities where they are abun-

dant in the diet. However, until safe exposure levels are determined, it is recommended that isoflavone supplementation be discontinued during pregnancy. SRCE is also not recommended for children under the age of two.

—*Risa N. Schulman, Ph.D.*

Enclosure: Copyright © 1998, Natural Products Research Consultants, Inc., reprinted with permission. All right reserved. For more information contact NPRC at tel. 973-540-4132 or fax 973-762-4504.

Bin #172