P.O. Box 144345 Austin, TX 78714-4345 ▪ 512.926.4900 ▪ Fax: 512.926.2345 ▪ www.herbalgram.org

# HerbClip™



**File: ■ Milk Thistle (*Silybum marianum*)**
**■ Silymarin Flavonolignans**
**■ Pharmacokinetics**

**HC 111363-493**

**Date: March 31, 2014**

**RE: Pharmacokinetics and Antioxidant Activity of Silymarin Flavonolignans**

Zhu H-J, Brinda BJ, Chavin KD, Bernstein HJ, Patrick KS, Markowitz JS. An assessment of pharmacokinetics and antioxidant activity of free silymarin flavonolignans in healthy volunteers: A dose escalation study. *Drug Metab Dispos.* September 2013;41(9):1679-1685.

This dose-response study investigated the pharmacokinetics of standardized silymarin in healthy subjects and assessed changes in oxidative status using the biomarker 8-epi-prostaglandin F2α (8-epi-PGF2α). The study material consisted of 175 mg of standardized milk thistle (*Silybum marianum*) fruit extract containing 140 mg of silymarin (Legalon® 140 mg; MADAUS GmbH; Cologne, Germany). Analysis showed that each capsule contained 21.2 mg of silybin A, 29.5 mg of silybin B, 11.4 mg of isosilybin A, 8.2 mg of isosilybin B, 31.5 mg of silychristin, 36.4 mg of silydianin, and 5.9 mg of taxifolin. In total, 14 healthy subjects were recruited for this study, underwent a physical examination, and provided their medical history. Blood and urine parameters were analyzed to determine health, and included subjects did not smoke, were not on medications or dietary supplements or vitamins, and were instructed to avoid grapefruit (*Citrus* × *paradisi*) juice, artichokes (*Cynara scolymus*) (to avoid dietary taxifolin consumption), caffeine, and alcohol 2 weeks before and during the trial.

This study took place at the Medical University of South Carolina, Charleston, South Carolina. After baseline fasting blood and urine collection, subjects were given 175 mg, 350 mg, or 525 mg of milk thistle extract along with water. Blood was obtained at baseline, 0.5, 1, 1.5, 2, 3, 4, 6, 8, 10, 12, and 24 hours. After a washout period of 7 days, the pharmacokinetic procedures were repeated 2 more times. A 28-day exposure to milk thistle extract (525 mg, taken as 175 mg 3 times daily) was also conducted for steady state analysis. Blood samples were taken at baseline prior to the first dosage of the day ($C_{trough}$) and at 0.5, 1, 2, 3, 4, and 8 hours following the dosages.

At 4 times during the 28-day dosage period, subjects were screened for adverse effects 2 hours after milk thistle extract dosage to align effects with maximum plasma concentration of compounds ($C_{max}$). Silymarin compounds were detected using liquid chromatography-mass spectrometry, and 8-epi-PGF2α was measured with gas chromatography-mass spectrometry. The pharmacokinetic parameters $C_{max}$, time to maximum plasma concentration ($T_{max}$), terminal elimination rate constant (λz), and elimination half-life ($t_{1/2}$) were determined. Additionally, the area under the curve (AUC) from time 0 to infinity for

single doses ($AUC_{0\rightarrow\infty}$) and from time 0 to 8 hours were calculated ($AUC_{0\rightarrow8}$), as well as the apparent clearance (CL/F) and the apparent volume distribution (V/F).

In total, 13 subjects aged 23-44 years old participated in the study; 1 subject violated the protocol and was dropped from the study. No adverse effects were reported in any subjects with either the single dose or 28-day dose experiments. The $C_{max}$ and $AUC_{0\text{-}24h}$ increased along with increasing single doses. The CL/F was not different between dosages. The $C_{max}$ of silybin A for the 175 mg dose was 106.9 ± 49.2 ng/ml, 200.5 ± 98 ng/ml for 350 mg of milk thistle extract, and 299.3 ± 101.7 ng/ml for the 525 mg dose. The $C_{max}$ of silybin B was lower than silybin A for all dosages (30.5 ± 16.3 ng/ml for 175 mg dose, 74.5 ± 45.7 ng/ml for 350 mg dose, and 121.0 ± 52.2 ng/ml for the 525 mg dose). The $C_{max}$ for isosilybin A was 6.1 ± 2.9 ng/ml, 18.2 ± 13.5 ng/ml, and 24.7 ± 11.8 ng/ml for 175 mg, 350 mg, and 525 mg doses, respectively, while the $C_{max}$ for isosilybin B was 22.0 ± 10.7 ng/ml, 46.4 ± 31 ng/ml, and 75.8 ± 32.3 ng/ml.

Although silychristin was not detected following the 175 mg dose, the $C_{max}$ was 4.6 ± 1.1 ng/ml and 8.5 ± 3.4 ng/ml for 350 mg and 525 mg doses. The $C_{max}$ for silydianin (6.5 ± 3.8 ng/ml) and taxifolin (5.1 ± 2.7 ng/ml) were only measurable with the 525 mg dose. The $T_{max}$ ranged from 1.0 to 1.5 hours for all compounds at all dosages. After the 28-day regimen of 525 mg daily of milk thistle extract, the pharmacokinetic parameters were as follows: $C_{max}$ of silybin A (134.7 ± 72.0 ng/ml, $T_{max}$ 2 hours), silybin B (42.1 ± 27.3 ng/ml, $T_{max}$ 1 hour), and isosilybin B (26.8 ± 19.9 ng/ml, $T_{max}$ 1 hour). A non-significant decrease was detected in 8-epi-PGF2α from baseline to endpoint (P=0.076). The concentrations of 8-epi-PGF2α were considerably decreased after 28 days but failed to reach statistical significance (P = 0.076).

In conclusion, the flavonolignans were rapidly absorbed and eliminated. Escalating single-dose assessments suggested dose proportionality. In order of abundance, the exposure to free (i.e., unconjugated) silymarin flavonolignans was greatest for silybin A followed by silybin B, isosilybin B, isosilybin A, silychristin, and silydianin. Stereoselective metabolism of silybin A and B and isosilybin A and B were also in evidence. Plasma concentrations of the flavonolignans were generally lower than those used in in vitro studies assessing various pharmacodynamic effects, although silybin A, silybin B, and isosilybin B reached systemic concentrations in the free form that could produce clinical effects. It is suggested that the decrease in 8-epi-PGF2α may be of greater magnitude in patients with chronic diseases associated with elevated oxidative stress, such as chronic liver diseases, as opposed to the healthy subjects in the present study. Furthermore, the milk thistle extract dosage of 175 mg 3 times daily may not be the optimal dosing regimen to achieve maximal antioxidant effects. It is suggested that the decrease in 8-epi-PGF2α, the marker of oxidative damage, has been muted in these healthy subjects, pointing to dosage considerations for patients with elevated oxidative damage. Further investigation is warranted to study antioxidant effects of silymarin extract in patients exhibiting elevated levels of oxidative stress.

*—Amy C. Keller, PhD*

The American Botanical Council has chosen not to include the original article.