Bioavailability of soybean isoflavones from aglycone and glucoside forms in American women[1–4]

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ABSTRACT

Background: Test results on the bioavailability of isoflavones in the aglycone or glucoside form in Eastern and Western human subjects are contradictory.

Objective: The objective was to investigate the bioavailability of the soy isoflavones daidzein and genistein in American women with typical American dietary habits after ingestion of the aglycone or glucoside form of isoflavones.

Design: Fifteen American women aged 46 ± 6 y participated in a randomized, double-blind study. Blood samples were collected 0, 1, 2, 4, 8, 12, 24, and 48 h after consumption of aglycone or glucoside tablets with breakfast. The plasma curves for daidzein, genistein, and equol were constructed and the postprandial maximum concentration (C_{max}), time to the maximum concentration (t_{max}), and area under the curve (AUC) were determined.

Results: Isoflavone concentrations peaked early (1–2 h) in plasma and peaked again at 4–8 h. Mean C_{max}, t_{max}, and AUC values for genistein were not significantly different after ingestion of aglycone or glucoside. However, C_{max} and AUC values, but not t_{max}, were significantly higher for daidzein after aglycone ingestion, which was partly due to its higher content in the aglycone tablets. Equol appeared after 4 h and remained elevated after 48 h. Despite a higher content of daidzein in the aglycone tablets, the AUC for equol was significantly higher after ingestion of the glucoside tablets, probably because of the metabolic action of intestinal bacteria during the long intestinal transit time of glucoside.

Conclusion: The apparent bioavailability of genistein and daidzein is not different when consumed as either aglycone or glucoside by American women. *Am J Clin Nutr* 2003;77:1459–65.

KEY WORDS Isoflavones, daidzein, genistein, equol, glucoside, aglycone

INTRODUCTION

Isoflavones, the major flavonoids of soybeans (1, 2), have recently gained great attention for their potential health benefits in the prevention of cancer, osteoporosis, postmenopausal syndromes, and hypercholesterolemia (1, 3–8). Isoflavones in soybeans are found in 4 chemical forms: aglycone, glucoside, acetylglucoside, and malonylglucoside; however, the glucoside forms are predominant (9), and variation in food processing techniques alter the relative content of acetylglucoside, malonylglucoside, and simple glucosides (10). In soybean foods, isoflavones exist in both the aglycone and glucoside forms; the fermentation process increases the aglycone forms in soy products. On average, cooked soybeans, texturized vegetable protein, and soy-milk powder contain >95% of the total isoflavones as glucoside, whereas soybean-fermented products such as tofu and tempeh contain ≈20% and ≈40%, respectively, of its isoflavones as aglycones (2). Isoflavone supplement products contain both forms of isoflavones. The major isoflavones in soybeans are daidzin (7,4′-dihydroxyisoflavone 7-glucoside) and genistin (4′,5,7-trihydroxyisoflavone 7-glucoside) as glycosides and their corresponding aglycone forms: daidzein (4′,7-dihydroxyisoflavone) and genistein (4′,5,7-trihydroxyisoflavone). To establish a relation between isoflavone intake and its proposed biological activity, the absorption, distribution, metabolism, and excretion of isoflavones from the glucoside and aglycone forms have been investigated in animals and humans (2, 11–18). After ingestion, the glucoside forms of isoflavones are hydrolyzed by β-glucosidases to the aglycone forms in the jejunum (19–21). The released aglycone forms of isoflavones are either absorbed intact by the intestine or further metabolized by intestinal microflora into several other products, including equol and *O*-desmethylangolensin (22–24) metabolites of daidzein and *p*-ethyl phenol metabolites of genistein before absorption (25). A study of Japanese women reported that isoflavone aglycones (a fermented soybean extract) were absorbed more efficiently than were isoflavone glucosides (an unfermented soybean extract) (18). In contrast, most recent studies reported that the bioavailability of daidzein and genistein isoflavones was greater when ingested as glucosides rather than as aglycones (26). In the latter study, plasma genistein concentrations were consistently higher than were daidzein concentrations when equal amounts of the 2 isoflavones were consumed.

TABLE 1
Composition of isoflavones in aglycone and glucoside tablets[1]

Isoflavone	Aglycone (259.2 mg)	Glucoside (250.6 mg)
	mg (mmol)	mg (mmol)
Daidzin	ND (ND)	6.18 (0.0148)
Glycitin	ND (ND)	1.02 (0.0023)
Genistin	ND (ND)	8.30 (0.0192)
Malonyl daidzin	ND (ND)	0.35 (0.0007)
Malonyl glycitin	ND (ND)	ND (ND)
Malonyl genistin	ND (ND)	0.53 (0.0010)
Acetyl daidzin	ND (ND)	0.39 (0.0008)
Acetyl glycitin	ND (ND)	ND (ND)
Acetyl genistin	ND (ND)	0.47 (0.0010)
Daidzein	5.28 (0.0208)	0.00 (0.0000)
Glycitein	0.73 (0.0026)	0.00 (0.0000)
Genistein	4.72 (0.0175)	0.00 (0.0000)
Total	10.72 (0.0408)	17.24 (0.0399)

[1]Composition analyzed by Kikkoman Corp, Noda, Japan. ND, not detectable.

This differential plasma concentration was accounted for by a larger volume of distribution of daidzein in the whole body compared with genistein. Hydrolysis of glucosides in isoflavone-rich drinks before ingestion showed that it did not improve the bioavailability of isoflavones in postmenopausal women (27).On the basis of animal studies, no differences in the absorption of isoflavones were found when glycoside and aglycone forms were consumed (12). However, results from human studies are inconclusive. Because the intestinal microflora, dietary habits, and exposure of American women to soy-based foods and products differ from those of Japanese women, in the present study we sought to investigate the postprandial absorption of the soy isoflavones daidzein and genistein after ingestion of aglycone and glucoside forms by American women.

SUBJECTS AND METHODS

Isoflavones

Two forms of soy isoflavones, aglycone and glucoside, were used in this study. The amount of daidzein and genistein and their derivatives in each aglycone and glucoside tablet is shown in **Table 1**. An aglycone tablet weighing 259.2 mg contained 0.0408 mmol isoflavones in aglycone form (10.72 mg), which was composed of 50.9% daidzein and 42.8% genistein and their derivatives (**Table 2**). A glucoside tablet weighing 250.6 mg contained 0.0399 mmol isoflavones in glucoside form (17.24 mg), which

TABLE 2
Isoflavone component ratio of glucoside and aglycone in isoflavone tablets[1]

Isoflavone	Aglycone (259.2 mg)	Glucoside (250.6 mg)
	mmol (mol%)	mmol (mol%)
Daidzein derivatives[2]	0.0208 (50.9)	0.0164 (41.1)
Glycitein derivatives	0.0026 (6.3)	0.0023 (5.7)
Genistein derivatives	0.0175 (42.8)	0.0212 (53.2)
Total	0.0408 (100)	0.0399 (100)

[1]Composition analyzed by Kikkoman Corp, Noda, Japan.
[2]Daidzin + malonyl daidzin + acetyl daidzin − daidzein.

TABLE 3
Total intake of isoflavones in each test

	Daidzein	Genistein
	mmol	
3 Aglycone tablets	0.0624	0.0525
3 Glucoside tablets	0.0492	0.0636

was composed of 41.1% daidzein and 53.2% genistein and their derivatives.

Subjects

A total of 16 women between the ages of 39 and 53 y ($\overline{x} \pm$ SD: 46 ± 6 y) participated in this study. The subjects were selected from a total of 39 volunteers who were initially prescreened and selected by phone interview from a total of 145 volunteers who responded to an announcement posted in the local media. The recruited volunteers were further screened about health status and dietary habits with the use of a dietary questionnaire. Of 39 recruited subjects, 6 were rejected because of abnormal laboratory blood results and 17 because they had a dietary habit of consuming soy products. The exclusion criteria included a body mass index (BMI; in kg/m^2) < 20 and > 32, a vegetarian dietary pattern, lactose intolerance, current smoking, diseases or disorders of the gastrointestinal tract, and hypertension. Volunteers with a history of cancer but not currently consuming a special diet or medication were accepted at prescreening. Volunteers were asked to abstain from alcohol during the study period. The selected subjects were in good health and had typical American dietary habits. The subjects were asked not to change their dietary habits, not to consume foods containing soy or soy products or isoflavones, and not to change their physical activity patterns during the 12-d period of the study. One subject was eliminated from the study because of an unusual plasma isoflavone profile. The study was conducted in the Metabolic Research Unit (MRU) of the Jean Mayer US Department of Agriculture Human Nutrition Research Center on Aging at Tufts University. The study protocol was reviewed and approved by the Human Investigation Review Committee of Tufts University and the New England Medical Center. Informed written consent was obtained from all volunteers.

Experimental design

The study was randomized, was double blind, and consisted of 2 phases. On day 1 of the first phase of the study, the subjects who had fasted overnight (14 h) were admitted to the MRU at 0730. A venous blood sample (0 time) was collected into EDTA-containing tubes. Within 30 min, a standard breakfast was completely consumed, and the subject was asked to swallow 3 tablets of one form (aglycone or glucoside) of the isoflavone tablets. The total amounts of daidzein and genistein consumed by ingestion of 3 aglycone and 3 glucoside tablets are shown in **Table 3**. Breakfast was composed of corn flakes, low-fat (1%) milk, orange juice, a banana, distilled water, and optional instant coffee or tea with sugar or artificial sweetener and with or without cream. Additional blood samples were obtained into EDTA-containing tubes 1, 2, 4, 8, and 12 h after ingestion of the tablets. Subjects remained at the MRU during the 12-h period; they consumed a standard lunch at noon after blood was drawn at 4 h and consumed dinner at 1700 after blood was drawn at 8 h. Lunch was composed of lettuce, sliced tomato, shredded carrot, 2 pieces of white bread, tuna, mayonnaise, a fresh apple, a small package of
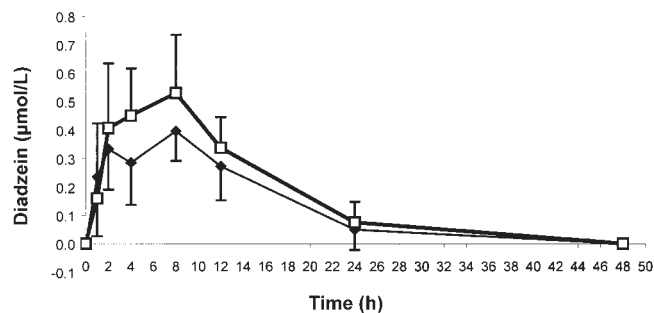
FIGURE 1. Mean (± SD) postprandial plasma concentrations of daidzein from glucoside (□) or aglycone (◆) in 15 women 0, 1, 2, 4, 8, 12, 24, and 48 h after a single intake of 3 tablets of isoflavone aglycone (a total of 0.0624 mmol daidzein) or 3 tablets of isoflavone glucoside (a total of 0.0429 mmol daidzein).
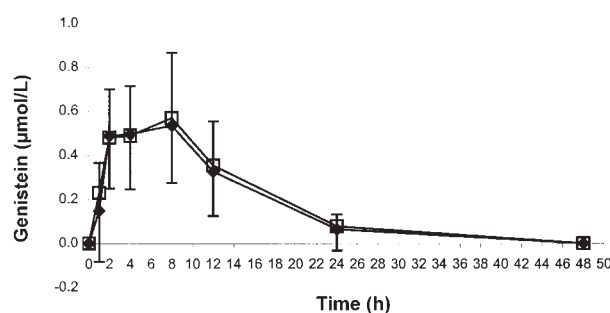

FIGURE 2. Mean (± SD) postprandial plasma concentrations of genistein from glucoside (□) or aglycone (◆) in 15 women 0, 1, 2, 4, 8, 12, 24, and 48 h after a single intake of 3 tablets of isoflavone aglycone (a total of 0.0525 mmol genistein) or 3 tablets of isoflavone glucoside (a total of 0.0636 mmol genistein).

butter cookies, a regular ginger ale, 8 g Italian or French dressing (optional), and salt and pepper. Dinner was composed of tomato soup, herbed chicken prepared with garlic powder, corn oil, oregano, cooking sherry, broccoli, a dinner roll, butter, vanilla ice cream, sparkling water, and salt and pepper (optional). The subjects spent the night at their own residences and were asked to return to the MRU to provide blood samples 24 and 48 h after ingestion of the tablets.

After a 6-d washout period, the same subjects were admitted to the MRU, and the same procedures were repeated in the second phase of the study. During this second phase of the study, the other form of isoflavone tablets was consumed at breakfast and was followed by blood sampling up to 48 h after ingestion, as described for phase 1. The order of tablet administration in each phase was randomized. The collected blood samples were spun at 2500 × g for 20 min at 4 °C within 40 min of blood being drawn and then the plasma was separated and stored under nitrogen gas at −70 °C until analyzed.

Measurement of plasma isoflavones

Plasma genistein and daidzein were measured by HPLC according to the method of Piskula et al (13). Fifty microliters of plasma were added to 50 μL acetate buffer (0.2 mol/L, pH 5.0) containing 500 U β-glucuronidase and 40 U sulfatase activity (Sigma Chemical Co, St Louis) and incubated for 1 h at 37 °C in a shaking water bath to release the free forms of isoflavones from the glucuronide and sulfate conjugates. Genistein and daidzein were extracted with 0.3 mL methanol:acetic acid (100:5, vol:vol) by being mixed by vortex and sonication, which was followed by centrifugation at 4000 × γ for 5 min at 4 °C. The supernatant fluid was diluted 1:0.5 (vol:vol) with the addition of 150 mmol lithium acetate/L (Sigma Chemical Co, St Louis), and an aliquot was injected into the HPLC column. The HPLC system consisted of a Waters 600E pump (Milford, MA), a Rheodyne 8125 injector (Rohnert Park, CA) with a 20-μL loop or Waters 717-plus auto sampler, a reversed-phase column (TSK-gel ODS-80TS QA column, 5 μm, 150 × 4.6 mm; Tosohaas, Montgomeryville, PA), a column temperature controller (Keystone Scientific, Bellefonte, PA), a BAS electrochemical detector (BAS, West Lafayette, IN), and a personal computer digital data station with Millenium32 software (version 3.05.01; Waters, Milford, MA). The composition of the mobile phase was water:methanol:acetic acid (57:41:2, vol:vol:vol) containing 50 mmol lithium acetate/L. The flow rate was set at 0.9 mL/min. The HPLC column was maintained at 40 °C and the eluent was mon-

itored for 35 min with an electrochemical detector set at +950 mV. The eluent of isoflavones was quantified by determining the peak areas in the chromatograms calibrated against known amounts of standards with high purity (> 99%). The lower limit of detection was 0.01 μmol/L for genistein and daidzein. All of the plasma samples collected from each subject during the 2 phases were analyzed in the same batch of analysis. The postprandial absorption and metabolism curves for genistein and daidzein were constructed by plotting the plasma concentrations over time. The postprandial maximum concentration in plasma (C_{max}) and the time for the maximum concentration to be reached (t_{max}) were determined from the curves, and apparent bioavailability was determined as the area under the curve (AUC) calculated by the trapezoidal method during the 12 and 48 h after ingestion of the tablets.

Statistical analysis

The data were statistically analyzed by paired Student's t test with the use of SYSTAT software (version 9.00; SPSS, Chicago). Data are presented as means ± SDs.

RESULTS

The group mean postprandial appearance and disappearance of daidzein and genistein in the plasma of women after ingestion of aglycone and glucoside forms are shown in **Figures 1** and **2**, respectively. There was an initial rapid increase in daidzein and genistein concentrations in plasma within 1–2 h of tablet consumption, followed by a plateau and then a second increase; C_{max} was reached at 4–8 h. Thereafter, the plasma concentration of isoflavones decreased at 12 and 24 h, after which they were not detectable at 48 h. The mean plasma C_{max} for daidzein after the consumption of 3 aglycone tablets (that provided a total of 0.0626 mmol daidzein) was 0.530 ± 0.205 μmol/L, which was significantly higher than the mean C_{max} of 0.396 ± 0.104 μmol/L after the consumption of 3 glucoside tablets, which contained less daidzein (total: 0.0492 mmol; Table 3). However, the mean plasma C_{max} for genistein after consumption of the aglycone tablets was not significantly different from that after consumption of the glucoside tablets (0.534 ± 0.333 compared with 0.569 ± 0.294 μmol/L, respectively), even though the genistein derivatives were higher in the glucoside tablets than in the aglycone tablets (0.0636 compared with 0.0525 mmol; Table 3).

The subjects responded differently after isoflavone consumption. The plasma t_{max} for the peak concentration of genistein and
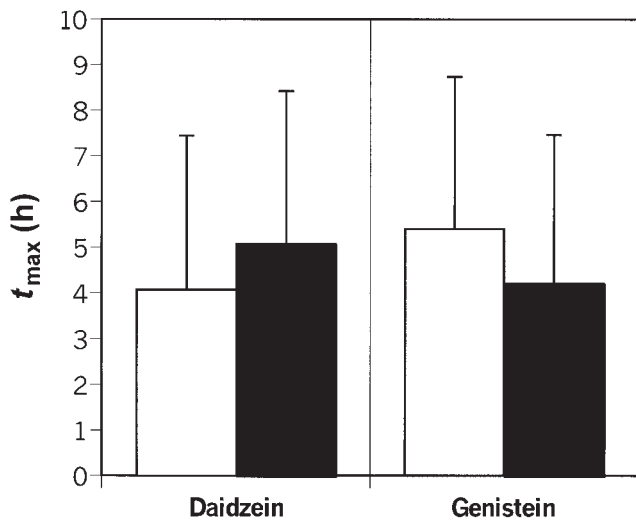
FIGURE 3. Mean (± SD) time required for daidzein and genistein to reach the maximum plasma concentration (t_{max}) in 15 women after intake of isoflavones as glucoside (□) or aglycone (■).

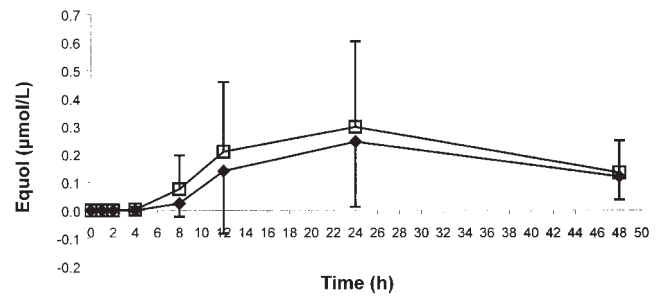**FIGURE 4.** Mean (± SD) postprandial plasma concentrations of equol, a metabolite of daidzein, in 15 women after intake of isoflavones as glucoside (□) or aglycone (◆).

daidzein was different between subjects. After an initial peak of isoflavones at 1–2 h, a second peak, although not in all of the subjects, appeared 8–12 h after intake of the isoflavone tablets.

After consumption of the glucoside tablets, 10 subjects had the initial daidzein peak at 1–2 h, 10 subjects had C_{max} at 8 h, and 5 subjects had only one daidzein peak (ie, no second peak). After consumption of the aglycone tablets, only 8 of 15 participants showed the initial peak of daidzein within 1–2 h, 9 subjects had C_{max} at 8 h, and 6 subjects had only a daidzein peak (one subject had t_{max} at 1 h, 4 subjects had t_{max} at 4 h, and 3 subjects had t_{max} at 8 h).

After consumption of the glucoside tablets, only 7 subjects showed an initial rapid rise of genistein in plasma within 1–2 h, with a subsequent C_{max} at 8 h. Although one subject showed only a single peak of genistein C_{max} at 1 h, 4 subjects had C_{max} at 4 h, and 3 subjects had C_{max} at 8 h. After consumption of the aglycone tablets, only 3 subjects had the initial plasma peak of genistein at

2 h, followed by C_{max} at 8 h. One subject had the initial peak of genistein in plasma at 4 h and C_{max} at 12 h. The 11 subjects who showed only one peak of genistein in plasma after consumption of aglycone had different t_{max} values (2–12 h).

The mean t_{max} values for plasma daidzein and genistein after consumption of the aglycone tablets were not significantly different from the values after consumption of the glucoside tablets (**Figure 3**).

Because there were considerable variations on the rate of absorption and appearance of isoflavones in plasma among the volunteers, the apparent bioavailability of daidzein and genistein from aglycone and glucoside sources was calculated as the AUC and is shown in **Table 4**. The bioavailability of daidzein from the aglycone form was ≈25% higher than the bioavailability of daidzein from the glucoside form (8.3 ± 2.6 compared with 6.2 ± 1.7 µmol · 48 h/L; P < 0.05), whereas the bioavailability of genistein was not significantly different from either the aglycone or glucoside forms (8.3 ± 4.2 compared with 8.9 ± 4.7 µmol · 48 h/L). Because all of the subjects consumed the same defined and controlled diet during their first 12 h of the absorption phase while they stayed at the MRU, we further compared the AUCs for plasma daidzein and genistein over a 12-h period. As shown in Table 4, the same bioavailability patterns were observed during a 12-h period as were observed during the 48-h period, ie, the bioavailability of daidzein from aglycone was higher than that from glucoside, whereas the bioavailability of genistein was not different from either form.

Because each volunteer received the same amount of isoflavones tablets in each phase, the plasma concentration might have been affected by the distribution volume of the analytes. Therefore, we adjusted the plasma concentration by BMI, which takes into account the distribution volume of daidzein and genistein. This adjustment did not change the results described above (Table 4).

Equol, one of daidzein's metabolites produced by the microflora of the intestine, is absorbed and appears in plasma after intake of daidzein and remains in plasma for a relatively longer period of time than do genistein and daidzein (22, 28). As shown in **Figure 4**, equol concentrations in plasma were negligible until 4 h, after which time they rose and reached a maximum concentration 24 h after the ingestion of the isoflavones; thereafter, equol concentrations in plasma decreased but remained higher than the baseline concentrations 48 h after ingestion. There was also a high variability in equol production among the subjects in this study. Therefore, the AUCs over 48 h were calculated and adjusted to

TABLE 4
Area under the curve (AUC) for plasma daidzein and genistein in women after a bolus intake of the isoflavones aglycone or glucoside[1]

	Intake	
	Glucoside	Aglycone
AUC for plasma daidzein		
(µmol · 48 h/L)	6.2 ± 1.7	8.3 ± 2.6[2]
(µmol · 12 h/L)	3.7 ± 0.9	4.9 ± 1.3[3]
(µmol · 48 h/L × BMI)	152 ± 34	203 ± 61[2]
(µmol · 12 h/L × BMI)	92 ± 22	121 ± 32[4]
AUC for plasma genistein		
(µmol · 48 h/L)	8.9 ± 4.7	8.3 ± 4.2
(µmol · 12 h/L)	5.4 ± 2.0	5.2 ± 2.2
(µmol · 48 h/L × BMI)	217 ± 103	201 ± 90
(µmol · 12 h/L × BMI)	132 ± 48	126 ± 48

[1] $\bar{x}$ ± SD; n = 15.
[2–4] Significantly different from glucoside (paired Student's t test): [2] P = 0.001, [3] P = 0.003, [4] P = 0.002.

TABLE 5
Area under the curve (AUC) of the plasma concentration of equol, a metabolite of daidzein, in women after a bolus intake of the isoflavones aglycone or glucoside[1]

	Intake	
	Glucoside	Aglycone
AUC for plasma equol		
(μmol·48 h/L)	9.0 ± 7.4	7.1 ± 5.6[2]
(μmol·48 h/L × BMI)	222 ± 183	174 ± 138[3]

[1] x ± SD; n = 15.
[2,3] Significantly different from glucoside (paired Student's t test): [2] $P = 0.04$, [3] $P = 0.03$.

BMI and are presented in **Table 5**. There were significantly higher plasma equol concentrations in subjects after ingestion of glucoside than after ingestion of aglycone over the 48-h period. This difference remained significant after adjustment for BMI.

DISCUSSION

The chemical forms in which isoflavones appear in food or supplements have been considered to be important for their bioavailability and thus for their biological activity. Glucoside conjugates of isoflavones are the major naturally occurring isoflavones in soybean and soybean-based food products (2, 29). The intestinal absorption of isoflavones requires a release of free forms from glucosidic conjugates. Thus, the intestinal absorption of glucoside forms of isoflavones has been thought to be delayed until they reach the large intestine, where the action of colonic microflora releases aglycones (30). However, this may not be important because, after ingestion, glucoside forms are hydrolyzed by β-glucosidases in the small intestine and appear in plasma within a short period of time (between 30 min and 2 h) (21, 31). Other factors—including dietary habits, the food matrix, the extent of intestinal bacterial fermentation, intestinal transit time, and age—may influence the intestinal metabolism and bioavailability of isoflavones in humans (7, 32).

In the present study, all the women were selected from a pool of volunteers after they were screened for dietary habits, BMI, and gastrointestinal function; all were in good health. It is important to note that at least for the 12-h period in each phase during which the subjects stayed at the MRU, all of the subjects consumed the defined diet at breakfast, lunch, and dinner. Thus, the food matrix, which may influence the bioavailability of isoflavones during the absorption period (7), was the same for all of the participants, and thus it is assumed that the appearance of daidzein, genistein, and equol in plasma is not influenced by the ingested food. However, there were considerable variations in the kinetics of the appearance and disappearance of isoflavones among the subjects, which may reflect the subjects' differences in intestinal microflora composition, past exposure to isoflavones, and glucosidase production. Some volunteers showed an early rise of genistein and daidzein within 1–2 h of tablet ingestion, followed by the second peak at 4–8 h, reflecting an enterohepatic circulation (7).

Overall, the form of isoflavones as aglycone or glucoside made no difference in the pattern of plasma appearance and disappearance of genistein. Furthermore, there was no difference in the bioavailability of genistein from the aglycone or glucoside form when they were consumed with a typical American diet by American women. It is evident from our findings that there was no difference in mean t_{max} and C_{max} values for genistein between the aglycone and glucoside forms. The absence of the difference in postprandial concentrations of genistein after consumption of the aglycone or glucoside form was apparent when the AUCs over 12- or 48-h periods were compared (Table 4). Furthermore, adjustment of the AUCs for the subjects' BMIs, which takes into account the volume of distribution of isoflavones, did not change the results. This lack of difference in the bioavailability of genistein between the aglycone and glucoside forms was present despite a 17% higher amount of genistein in the total of 3 glucoside tablets (0.0636 mmol) than in the total of 3 aglycone tablets (0.0525 mmol) (Table 3).

In contrast, we found a significantly higher C_{max} for plasma daidzein and a larger AUC for plasma daidzein 12 and 48 h after ingestion of the aglycone form than after ingestion of the glucoside form. These differences of ≈24% remained significant after adjustment for BMI (Table 4). It is important to note that this difference in the bioavailability of daidzein from the aglycone form can be attributed in part to the 21% higher amount of daidzein (0.0624 mmol) in the total of 3 aglycone tablets than in the total of 3 glucoside tablets (0.0492 mmol) (Table 3). Compared with the aglycone form, the glucoside form may stay in the intestines for a longer time and be subjected to both bacterial metabolism and intestinal glucosidase enzymes. Nevertheless, there was no difference between aglycone and glucoside on the t_{max} for plasma daidzein.

There are conflicting results in the literature on the bioavailability of isoflavones when consumed in the aglycone form compared with when consumed in the glucoside form. Our observation agrees with some reports indicating that the bioavailability of genistein and daidzein is not influenced by the presence of free or conjugated forms in the diet or in the supplements (27). Richelle et al (27) reported that isoflavone-rich extract, which was enzymatically hydrolyzed by commercial glucosidase to produce aglycones, had the same plasma pharmacokinetics over a 34-h period in postmenopausal women as after ingestion of an isoflavone-rich extract drink without hydrolysis (glucoside form). Setchell et al (26) reported that even though the plasma t_{max} of daidzein and genistein was shorter when the aglycone form was consumed, the bioavailability of daidzein and genistein, as determined from the AUC of the plasma concentration of these compounds, is higher after consumption of the glucoside than after the aglycone form (7, 26). It was suggested by Setchell et al (26) that a greater bioavailability of isoflavones from the glucoside form is due to the glucoside moiety acting as a protecting group in the molecule to prevent biodegradation of the isoflavone structure.

Our results are in contrast with those of Izumi et al (18), who reported that the bioavailability of both genistein and daidzein from the aglycone form was significantly higher and faster than that from the glucoside form when consumed by 8 Japanese volunteers. This difference was observable with either low or high bolus doses of isoflavone. A variety of factors may contribute to the differential results, including ethnic background, dietary habit, intestinal microflora, food matrix, the administered dose, and the number of subjects in a study.

In the present study, the participating subjects were all Americans with typical American dietary habits. Thus, it is plausible that participants in this study had a somewhat different intestinal bacterial composition than did Japanese men and women in Izumi et al's study (18), who might have had dietary habits that were different from those of American women. It is also possible that

American women, whose colonic microfloral populations differ from those of Japanese women, may have developed a higher capacity to hydrolyze glucosides in the intestines than did the Japanese women. Thus, the American women may have metabolized the glucoside forms of isoflavones more efficiently than did the Japanese women. The differences in the intestinal microfloral populations between the Japanese women who consumed the traditional Japanese diet and the women who consumed a typical Western diet was noted previously (22). In food-deprived animals, the absorption of isoflavones was shown to be significantly higher and faster than in fed animals (13); thus, the presence of food and a potentially different food matrix may influence the bioavailability of isoflavones from the gastrointestinal tract. In Izumi et al's study, Japanese volunteers consumed the isoflavone tablets after a breakfast consisting of 2 onigiri (rice balls) and green tea (personal communication), whereas in the present study the participating subjects consumed the isoflavone tablets after a breakfast composed of corn flakes cereal, banana, orange juice, and milk with or without instant tea or coffee.

Furthermore, the number of subjects who participated in our study was greater (15 subjects) and all of the participants were female as opposed to the participants in Izumi et al's study (18) and the other abovementioned studies, who were fewer in number (6–8 subjects) and were of both sexes.

We observed that equol, a bacterial metabolite of daidzein produced in the intestines, appeared in the plasma of subjects ≈4 h after ingestion of the isoflavone tablets. The apparent bioavailability of equol over the 48 h after ingestion, ie, the AUC, was higher after consumption of the glucoside tablets than after consumption of the aglycone tablets despite the higher concentration of daidzein in the aglycone than in the glucoside tablets. This observation leads us to speculate that one possible reason for the lower concentrations of daidzein in plasma (AUC) after the ingestion of glucoside than after the ingestion of aglycone is in part related to the bacterial metabolic conversion of daidzein to equol in the intestines because of a longer transit time for glucosides than for aglycones (22, 23). In summary, the bioavailability of genistein and daidzein, the major isoflavones in soy and soy products, is not significantly different when the isoflavones are consumed as either aglycone or glucoside by American women with typical American dietary habits.

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