Whole Sesame Seed Is as Rich a Source of Mammalian Lignan Precursors as Whole Flaxseed

Karen D. Coulman, Zhen Liu, Winston Quan Hum, John Michaelides, and Lilian U. Thompson

Abstract: The mammalian lignans enterolactone and enterodiol, which are produced by the microflora in the colon of humans and animals from precursors in foods, have been suggested to have potential anticancer effects. This study determined the production of mammalian lignans from precursors in food bars containing 25 g unground whole flaxseed (FB), sesame seed (SB), or their combination (FSB; 12.5 g each). In a randomized crossover study, healthy postmenopausal women supplemented their diets with the bars for 4 wk each separated by 4-wk washout periods, and urinary mammalian lignan excretion was measured at baseline and after 4 wk as a marker of mammalian lignan production. Results showed an increase with all treatments (65.1–81.0 μmol/day; P < 0.0001), which did not differ among treatments. Lignan excretion with the whole flaxseed was similar to results of other studies using ground flaxseed. An unidentified lignan metabolite was detected after consumption of SB and FSB but not of FB. Thus, we demonstrated for the first time that 1) precursors from unground whole flaxseed and sesame seed are converted by the bacterial flora in the colon to mammalian lignans and 2) sesame seed, alone and in combination with flaxseed, produces mammalian lignans equivalent to those obtained from flaxseed alone.

Introduction

The mammalian lignans enterolactone (EL) and enterodiol (ED) are produced by the action of bacterial flora in the colon of humans and animals on precursors in foods (Fig. 1) (1,2). Although the plant lignans secoisolariciresinol (SECO) diglucoside (SDG) and matairesinol are originally thought to be the only precursors of mammalian lignans (1,2), recent studies have shown that pinoresinol, lariciresinol, syringaresinol, arctigenin, and 7-hydroxymatairesinol can also be converted to these compounds (3,4). Once formed, the mammalian lignans are absorbed, circulating enterohepatically, and, in part excreted in the urine (5). Hence, consumption of foods rich in the mammalian lignan precursors has been shown to greatly increase urinary ED and EL concentrations, and urinary lignan excretion has been used as a marker of the intake levels of lignan-rich foods (6–12).

Mammalian lignans are of interest because they have been suggested to have anticancer effects. Because of their chemical structural similarity to estrogen and their ability to bind to estrogen receptors α and β (13), they are thought to have weak estrogenic/antiestrogenic properties and thus can reduce the risk of hormone-related diseases such as breast cancer. Some epidemiological studies showed a negative relationship between breast cancer risk and the plasma levels, urinary excretion, or intake of lignans (14–18), although some studies also showed no relationship (19–21). Treatment with the mammalian lignan precursors SDG (22–25) and hydroxymatairesinol (4) or with the mammalian lignan EL (26) reduced mammary tumor development in carcinogen-treated rats. SDG also reduced the metastasis of estrogen receptor-negative human tumors (M3A-MB-435) (27,28) and of melanoma cells (29). In in vitro studies, mammalian lignans inhibited the activity of enzymes involved in hormone biosynthesis (aromatase, 7β-hydroxysteroid dehydrogenase, and 5α-reductase) (30–33) and the growth-stimulating effect of estrogen (34). They have antioxidant (35,36) and antiangiogenesis (37) properties and can reduce the adhesion, migration, and invasion of MDA-MB-435 cells, steps that are involved in the metastasis process (38).

Flaxseed has been reported to be one of the richest sources of mammalian lignan precursors, particularly SDG, with levels 100–800 times higher than that of other plant foods (39,40). Therefore, it has been used as a food model to determine the anticancer effects of lignans, the results of which have recently been reviewed (41). It reduced mammary tumor development when given at the preinitiation, promotion, or late promotion stages of carcinogenesis in carcinogen-treated rats (23,24,41,42). It also reduced the tumor growth and metastasis of estrogen receptor-negative human...
breast cancer cells (MDA-MB-435) (27,28,43,44) and the tumor growth of estrogen receptor-positive human breast cancer cells (MCF-7) in athymic mice (45). Reduced cell proliferation and erbB2 expression and increased cell apoptosis were also observed in the tumors of breast cancer patients who consumed flaxseed (46). Because the effect of SDG at levels found in flaxseed was similar to that of flaxseed, the anticancer effects of flaxseed have been attributed largely to the mammalian lignans produced from its SDG and other precursors (23–25,27,28,41).

Sesame seed is an oilseed that contains high concentrations of the plant lignans sesamin (SES), sesamolin (SMN), and sesaminol (Fig. 1) (47). It is unknown whether these lignans are converted to ED and EL because they are primarily fat soluble in contrast to the water-soluble SDG in flaxseed.

Many studies on sesame seed and its lignans have concentrated on their antioxidant (48–50) and blood pressure- (51–53) and serum lipid-lowering effects (50,54–58). Only limited studies have been conducted on their potential anticancer effects. Dietary SES has been shown to reduce the number of mammary tumors in carcinogen-induced rats (59). In vitro studies, SES, sesaminol, and SMN inhibited the growth of human lymphoid leukemia Molt 4B cells by inducing apoptosis (60–62). Sesamol, a sesaminol derivative produced from sesame oil refining or heat treatment, also induced growth arrest and apoptosis in MCF-7 and T47D breast cancer cells (63). In epidemiological studies, sesame oil-containing lignans were associated with decreased risk of stomach cancer (64).

The previous studies on flaxseed and sesame seed have been conducted using the ground seed or pure lignans, and it is unknown whether the production of mammalian lignans in unground whole seed found in many commercially available food products would be the same as that in the ground seed. There is a concern that the hard seed coat, particularly of flaxseed, prevents the release and metabolism of the seed components if they are eaten unground, thereby lessening their physiological effects. There is also a lack of human studies examining both the production of mammalian lignans and the physiological effects of ground or unground sesame seed. The combination of flaxseed and sesame seed has also...
not been studied. Because the two seeds contain different types of lignans, which may complement each other, they may have better health benefits when consumed together than when taken alone. Thus, the objective of this study was to examine, in postmenopausal women, the production of mammalian lignans from precursors in food bars containing 25 g unground flaxseed, sesame seed, or their combination. If they are produced, the whole seeds in bar formulation can be used in future studies to further demonstrate in clinical studies the potential anticancer effects of flaxseed and sesame seed alone or in combination.

Subjects and Methods

Subjects

Postmenopausal women were chosen as subjects for this study because many breast cancer patients are postmenopausal and may also consume foods rich in phytoestrogens such as lignans as potential alternative or complementary treatments for menopausal symptoms, osteoporosis, and cancer (65). Sixteen healthy, postmenopausal women (mean age 66.4 ± 1.7 yr; 14.4 ± 2.0 yr since menopause) volunteered in this study and nobody dropped out. At the commencement of the study (first baseline measurement), the mean weight and body mass index of the women were 73.1 ± 3.1 kg and 28.6 ± 1.5 kg/m², respectively. Both remained constant throughout the duration of the study. Subjects were included if they were omnivorous nonsmokers with stable body weights. Subjects were excluded if they were taking or had ever taken hormone replacement therapy, antibiotics within the last 6 mo, and/or lipid-lowering drugs 4 wk prior to beginning the study. The consumption of food containing flaxseed, sesame seed, or soy and the use of herbal supplements were discontinued 4 wk prior to the commencement of the study. Basic vitamin and mineral supplements, if taken regularly before commencing the study, were allowed to continue for the duration of the study. All subjects provided written informed consent, and the study protocol was reviewed and approved by the University of Toronto Human Ethics Committee.

Study Design

In a randomized crossover design, subjects supplemented their normal diets with one of three treatment food bars—flaxseed bar (FB), sesame seed bar (SB), or flaxseed+sesame seed bar (FSB)—for a period of 4 wk with a 4-wk washout period between each treatment. Subjects were randomized to the order in which they received FB, SB, and FSB. Similar to our previous study on flaxseed (11), 2 days prior to the beginning of the study (pre-study Day 1), subjects consumed a standard low-fiber, low-lignan dinner. On the morning of the day prior to the beginning of the study (pre-study Day 2), subjects collected a 24-h urine sample (first urine of day discarded) through to the first urine of Day 1 of the study period while continuing to consume standard low-fiber meals. The first treatment bar was then consumed at breakfast on study Day 1 and every day at breakfast after that up until Day 28. On the evening prior to the last treatment bar consumption (Day 27), subjects again consumed a standard dinner and began collecting a 24-h urine sample starting on the morning of Day 28 through to Day 1 of the next 4-wk period as previously described for pre-study Day 2 to study Day 1. Subjects ate their normal diets throughout the rest of the study days (Day 1–26); however, they were required to record 3 days of food intake (one weekend, two weekdays) during each treatment and washout periods. Subjects avoided other flaxseed-, sesame seed-, or soy-containing foods and herbal supplements throughout the course of the study. Macronutrient intakes from the diet records were calculated and averaged using the NutriWatch nutrient analysis program (version 6.1.22E Delphi 1, based on the 1997 Canadian Nutrient File; Elizabeth Warwick, PEI, Cornwall, Canada).

Food Bar Formulation and Analysis

Each bar contained 25 g flaxseed (Pizzey’s Milling, Angusville, MB, Canada), 25 g sesame seed (Grain Process Enterprises, Ltd., Scarborough, ON, Canada), or 12.5 g flaxseed and 12.5 g sesame seed plus 6.7 g high-fructose corn syrup and 5.6 g sugar to allow formulation into bar form and provide flavor. The ingredients were blended, portioned into bar format, and heated for 8 min at 190°C in a conventional oven. The bars were then cooled at room temperature for at least 60 min, vacuum-sealed in polyethylene film, and stored at −20°C until distributed to subjects. Subjects kept the bars in the freezer until the day of consumption.

Protein, carbohydrate, fat, and total, insoluble, and soluble dietary fiber contents of food bars were analyzed according to the standard methods of the Association of Official Analytical Chemists (66). The plant lignans (SDG, matairesinol, laricresinol, pinoresinol, sesaminol, SES, and SMN) in the treatment bars were analyzed by capillary gas chromatography-mass spectrometry (GC-MS) using a modification of other methods (7,39,67). Briefly, the samples were ground and defatted with hexane, and the lignans from the defatted ground samples were extracted with 70% MeOH, hydrolyzed with NaOH, and then neutralized with acetic acid. The lignan-rich fraction was eluted using C18 reversed-phase octadecylsilane bonded silica (C18) extraction columns (Scientific Products and Equipment, Ltd., Concord, ON), dried, and redissolved in sodium acetate buffer. After overnight enzyme hydrolysis with β-glucuronidase (Helix pomatia; Sigma Chemical Co., St. Louis, MO), samples were again passed through a C18 column, dried, and redissolved in MeOH. The internal standards 5α-androstan-3β,17β-diol and stigmasterol (Steraloids, Inc., Wilton, NH) were added, MeOH was evaporated under nitrogen, and the lignans were derivatized (Tri-Sil Reagent; Pierce, Rockford, IL), dissolved in hexane, and analyzed by GC-MS (GC 6890 Series
II, MS 5973; Hewlett-Packard, Avondale, PA) as we have previously described (7,8,39).

Because sesame lignans are primarily present in oil, their levels were also determined in the oil extracted from the seed by hexane. After removal of hexane from the extract by rotary evaporator, the pure oil (0.04 g) was redissolved in hexane (1 ml). To an aliquot (40 µl) was then added 40 µl of internal standard (1,000 µg/ml of 4′-OH-flavanone) and 120 µl methanol. After vortex mixing and filtration through a 0.45-µm pore-size filter, an aliquot (10 µl) of the filtrate was injected into a high-performance liquid chromatograph (Waters 2690 separation system; Milford, WA) coupled to a Waters 996 photodiode array detector equipped with a Waters symmetry C18 column (4.6 × 75 mm, 3.5 µm). Elution was with a gradient solvent system containing solvent A (water) and solvent B (acetonitrile) programmed at 20% B to 50% B in 15 min, 50% B to 100% B in 15 min, and then 20% B in 5 min running at a flow rate of 1 ml/min. The total lignan in the sample was the sum of the levels present in the defatted sample and the extracted oil.

Urinary Lignan Analysis

The concentrations of lignans [for example, ED, EL, and SECO (aglycone of SDG)] were measured in the urine by a GC-MS procedure, which is routinely used in our laboratory as previously described (7,8,11,39).

Statistical Analyses

All statistical analyses were performed using PROC MIXED, version 8.1 (SAS Institute, Cary, NC). Urinary lignans were corrected for order, treatment, and order × treatment. Least-squares means was performed to detect if treatments resulted in any significant change between baseline and follow-up measurements. Tukey-adjusted pairwise comparisons were performed to detect any differences between changes in treatments. The acceptable level of significance was $P < 0.05$.

Results

Food Bar Composition

Table 1. Composition of Treatment Bars With 25 g Seed

<table>
<thead>
<tr>
<th>Macronutrient</th>
<th>FB</th>
<th>SB</th>
<th>FSB</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total energy (kJ)</td>
<td>685</td>
<td>878</td>
<td>776</td>
</tr>
<tr>
<td>Protein (g, % energy)</td>
<td>5.2 (12.7)</td>
<td>6.0 (11.4)</td>
<td>5.6 (12.1)</td>
</tr>
<tr>
<td>Fat (g, % energy)</td>
<td>11.1 (60.7)</td>
<td>15.9 (68.2)</td>
<td>13.4 (64.7)</td>
</tr>
<tr>
<td>Available CHO (g, % energy)</td>
<td>10.9 (26.6)</td>
<td>10.7 (20.4)</td>
<td>10.8 (23.2)</td>
</tr>
<tr>
<td>Total dietary fiber (g)</td>
<td>7.0</td>
<td>3.1</td>
<td>4.7</td>
</tr>
<tr>
<td>Insoluble fiber</td>
<td>4.4</td>
<td>3.0</td>
<td>4.2</td>
</tr>
<tr>
<td>Soluble fiber</td>
<td>2.6</td>
<td>0.1</td>
<td>0.5</td>
</tr>
<tr>
<td>SDG (µmol)</td>
<td>200.05</td>
<td>0.04</td>
<td>100.04</td>
</tr>
<tr>
<td>Matairesinol (µmol)</td>
<td>0.64</td>
<td>0.08</td>
<td>0.36</td>
</tr>
<tr>
<td>Pinoresinol (µmol)</td>
<td>2.47</td>
<td>5.58</td>
<td>4.02</td>
</tr>
<tr>
<td>Lariciresinol (µmol)</td>
<td>1.75</td>
<td>0.21</td>
<td>0.98</td>
</tr>
<tr>
<td>Sesamin (µmol)</td>
<td>0</td>
<td>378.88</td>
<td>189.44</td>
</tr>
<tr>
<td>Sesamolin (µmol)</td>
<td>0</td>
<td>90.44</td>
<td>45.22</td>
</tr>
<tr>
<td>Sesaminol (µmol)</td>
<td>0</td>
<td>69.37</td>
<td>34.69</td>
</tr>
</tbody>
</table>

a: Abbreviations are as follows: FB, flaxseed bar; SB, sesame seed bar; FSB, flaxseed + sesame seed bar; CHO, carbohydrate; and SDG, secoisolariciresinol diglycoside.

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Table 2. Macronutrient Intake of Subjects

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Energy (kJ)</th>
<th>Protein (g)</th>
<th>CHO (g)</th>
<th>Dietary Fiber (g)</th>
<th>Fat (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baseline</td>
<td>7,056 ± 318</td>
<td>71.2 ± 2.8</td>
<td>225.2 ± 9.7</td>
<td>19.5 ± 1.4</td>
<td>56.0 ± 5.3</td>
</tr>
<tr>
<td>FB</td>
<td>7,629 ± 347</td>
<td>81.8 ± 4.7</td>
<td>232.9 ± 10.5</td>
<td>26.2 ± 1.7</td>
<td>64.4 ± 6.1</td>
</tr>
<tr>
<td>Change</td>
<td>573 ± 331</td>
<td>10.6 ± 3.9b</td>
<td>7.7 ± 10.3</td>
<td>6.7 ± 1.2c</td>
<td>8.4 ± 5.0</td>
</tr>
<tr>
<td>Baseline</td>
<td>6,868 ± 314</td>
<td>70.5 ± 3.5</td>
<td>228.6 ± 11.1</td>
<td>19.9 ± 1.6</td>
<td>51.9 ± 5.6</td>
</tr>
<tr>
<td>SB</td>
<td>7,909 ± 414</td>
<td>79.9 ± 4.2</td>
<td>237.5 ± 11.9</td>
<td>23.2 ± 1.7</td>
<td>73.5 ± 5.8</td>
</tr>
<tr>
<td>Change</td>
<td>1,041 ± 367d</td>
<td>9.4 ± 3.9b</td>
<td>8.9 ± 8.6</td>
<td>3.3 ± 1.4d</td>
<td>21.6 ± 6.3c</td>
</tr>
<tr>
<td>Baseline</td>
<td>7,378 ± 372</td>
<td>74.3 ± 3.3</td>
<td>229.0 ± 7.7</td>
<td>21.0 ± 1.4</td>
<td>62.6 ± 8.3</td>
</tr>
<tr>
<td>FSB</td>
<td>7,603 ± 368</td>
<td>76.8 ± 4.3</td>
<td>232.6 ± 14.0</td>
<td>23.9 ± 1.6</td>
<td>65.6 ± 5.0</td>
</tr>
<tr>
<td>Change</td>
<td>225 ± 470</td>
<td>2.5 ± 3.7</td>
<td>3.6 ± 13.5</td>
<td>2.9 ± 1.2b</td>
<td>3.0 ± 7.9</td>
</tr>
</tbody>
</table>

a: Abbreviations are as follows: CHO, total carbohydrate; FB, flaxseed bar; SB, sesame seed bar; and FSB, flaxseed + sesame seed bar. Values are expressed as mean ± SE. Change values in the same column are not significantly different at P > 0.05.

b: Change from baseline significant at P < 0.05.

c: Change from baseline significant at P < 0.001.

d: Change from baseline significant at P < 0.01.

Figure 2. Urinary lignan excretion of individual subjects pre- and posttreatment bars.
Another compound, which is excreted in the urine, is suggested by the presence of large amounts of mammalian lignans in bars with unground seed when compared with bars with ground seed. However, the urinary ED+EL excretion of this unknown compound was lower in the FSB group than can be predicted from those in the FB and SB groups. The urinary level of this compound increased significantly (P < 0.05) with SB and FSB, with the increase with SB (17.9 ± 3.5 µmol/day) significantly greater than that with FSB (6.4 ± 1.6 µmol/day) significantly greater, increase in urinary lignans than FB. This could not be predicted from those in the FB and SB groups.

**Discussion**

This study demonstrated for the first time that, in postmenopausal women, 1) plant lignans in unground whole flaxseed, sesame seed, and their combination, in the bar formulation, are converted by the bacterial flora in the colon to mammalian lignans, as indicated by the significantly large increase in urinary excretion of ED and EL; 2) large amounts of urinary ED and EL are produced from unground seed-containing bars, as much as that produced with flaxseed, indicating the presence of large amounts of mammalian lignan precursors; and 3) sesame plant lignans are converted as well to mammalian lignans from bars with unground seed with bars with ground seed. However, the urinary ED+EL excretion (62.2 µmol/day) after FB intake was higher than the 41.1 µmol/day observed in another study in which postmenopausal women also consumed 25 g/day ground flaxseed for 16 wk (8) and the 54.5 µmol/day excreted by premenopausal women who supplemented their diets with 25 g/day ground flaxseed for 7 days (11). In contrast, urinary ED+EL excretion of 59.4 µmol/day was reported in postmenopausal women after consuming 10 g/day ground flaxseed for 7 wk (9).

It is not possible to accurately compare the results of different studies as different subjects and flaxseed with perhaps different SDG concentrations were used (68). Differences in analytical methods to measure SDG and mammalian lignans can also produce different values (6). Subject differences in colonic microflora, intestinal transit time, or redox level of the colon (1,6) can also influence lignan metabolism, a fact that is also evident in this study. Subjects in the study (9), which produced higher urinary lignans per gram flaxseed intake, may have been greater mammalian lignan producers than the subjects in the present study. Nevertheless, despite individual variabilities, large urinary mammalian lignans observed in this study indicate that the bacterial conversion of the precursors in the whole seeds to mammalian lignans have taken place.

EL was produced in greater amounts than ED (53.2 vs. 9.0 µmol/day) in this study, which agrees with the results of Hutchins et al. (9) (56.2 vs. 3.2 µmol/day) and Brooks et al. (8) (31.1 vs. 10.0 µmol/day) in postmenopausal women but disagrees with the results of Nesbitt et al. (11) (10.9 vs. 43.6 µmol/day) and Cunnane et al. (69) (12.4 vs. 18.7 µmol/day) in premenopausal women. EL has been reported to possess greater beneficial effects than ED (38,70–72). Taken together, it would appear that flaxseed consumption leads to a greater excretion of EL in postmenopausal women and of ED in premenopausal women. However, more flaxseed studies are needed to confirm this as the differences could simply be due to differences in bacterial activities on the lignans independent of menopausal status. Known precursors of ED and EL such as SDG, matairesinol, pinonesinol, and lariciresinol were found in SB but at much lower concentrations, particularly in SDG, when compared with those in FB. It was thus surprising that SB consumption resulted in a greater, albeit not significantly greater, increase in urinary lignans than FB. This could not

<table>
<thead>
<tr>
<th>Treatment</th>
<th>SECO</th>
<th>ED</th>
<th>EL</th>
<th>ED+EL</th>
<th>Total (SECO+ED+EL)</th>
<th>Unknown Compound</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baseline</td>
<td>0.0 ± 0.0</td>
<td>0.2 ± 0.1</td>
<td>2.5 ± 0.6</td>
<td>2.7 ± 0.6</td>
<td>2.7 ± 0.6</td>
<td>0.0 ± 0.0</td>
</tr>
<tr>
<td>FB</td>
<td>2.9 ± 1.4</td>
<td>9.2 ± 1.7</td>
<td>55.7 ± 13.2</td>
<td>65.0 ± 14.0</td>
<td>67.8 ± 13.8</td>
<td>0.0 ± 0.0</td>
</tr>
<tr>
<td>Change</td>
<td>0.0 ± 0.0</td>
<td>0.2 ± 0.1</td>
<td>1.9 ± 0.4</td>
<td>2.1 ± 0.4</td>
<td>2.1 ± 0.4</td>
<td>0.0 ± 0.0</td>
</tr>
<tr>
<td>Baseline</td>
<td>0.4 ± 0.2</td>
<td>21.0 ± 6.2</td>
<td>61.7 ± 12.2</td>
<td>82.7 ± 12.9</td>
<td>83.1 ± 12.9</td>
<td>17.9 ± 3.5</td>
</tr>
<tr>
<td>SB</td>
<td>0.3 ± 0.2</td>
<td>20.8 ± 6.2</td>
<td>59.8 ± 12.2</td>
<td>80.6 ± 12.8</td>
<td>81.0 ± 12.9</td>
<td>17.9 ± 3.5</td>
</tr>
<tr>
<td>Change</td>
<td>0.3 ± 0.2</td>
<td>0.8 ± 0.7</td>
<td>4.2 ± 2.1</td>
<td>5.0 ± 2.8</td>
<td>5.3 ± 2.8</td>
<td>0.0 ± 0.0</td>
</tr>
<tr>
<td>FSb</td>
<td>8.2 ± 0.9</td>
<td>12.3 ± 3.1</td>
<td>51.2 ± 8.5</td>
<td>63.5 ± 10.1</td>
<td>71.7 ± 9.6</td>
<td>6.4 ± 1.6</td>
</tr>
<tr>
<td>Change</td>
<td>7.9 ± 3.7</td>
<td>11.5 ± 2.5</td>
<td>47.0 ± 8.1</td>
<td>58.5 ± 9.0</td>
<td>66.4 ± 9.2</td>
<td>6.4 ± 1.6</td>
</tr>
</tbody>
</table>

Table 3. Urinary Lignan Excretion Pre- and Posttreatment Bars

a: Values are mean ± SE; µmol/day. Abbreviations are as follows: SECO, secoisolariciresinol; ED, enterodiol; and EL, enterolactone; FB, flaxseed bar; SB, sesame seed bar; FSB, flaxseed + sesame seed bar.

b: Values are means ± SE; µmol equivalent internal standard/day; internal standard was 5α-androstane-3β,17β-diol. Changes in the same column with different superscripts (*, †, ‡) are significantly different at P < 0.05.
c: P < 0.0001 change from baseline.

d: P < 0.05 change from baseline.

Values are mean ± SE; µmol equivalent internal standard/day; internal standard was 5α-androstane-3β,17β-diol. Changes in the same column with different superscripts (*, †, ‡) are significantly different at P < 0.05.

c: P < 0.0001 change from baseline.
Figure 3. Gas chromatography-mass spectrometry chromatograms of urinary lignans after treatment bar consumption. IS1, 5α-androstane-3β,17β-diol (internal standard 1); 1, enterodiol; 2, enterolactone; 3, secoisolariciresinol; 4, compound A; and IS2, stigmasterol (internal standard 2).

Figure 4. Typical mass spectrum of unknown urinary compound A detected after consumption of sesame seed bar or flaxseed + sesame seed bar.
have been due to foods rich in mammalian lignan precursors consumed in the regular diet of the SB and FSB groups because the subjects did not consume flaxseed, soybean, and herbal products other than those in the treatment bars. Although whole grains have higher amounts of mammalian lignan precursors compared with refined grains (39, 40, 73), they were not consumed in large amounts by the subjects as seen in the diet record. The recommended level of dietary fiber Canadians should aim for in the diet is 25–30 g/day (74); however, our subjects’ baseline dietary fiber intake was only an average of 20.1 g/day. The small increase in dietary fiber intake during the study period can be attributed primarily to the food bar supplements. Furthermore, the twofold increase in urinary lignan excretion observed in healthy women who consumed 162 g whole meal rye bread compared with 153 g white wheat bread (73) is much smaller than the greater-than-eightfold increase in urinary lignan excretion after the flaxseed and/or sesame seed. Evidently, there are other mammalian lignan precursors in sesame seed, which may include its large amounts of SES, SMN, and sesaminol. These compounds have not previously been shown to be converted to the mammalian lignans. However, Nakai et al. (75) recently showed in rat studies that SES can be absorbed, metabolized in the liver to at least four compounds with catechol moieties, and excreted in bile and presumed also in the urine. The mass spectra of the unknown compound (compound A) detected in the urine of subjects after the intake of sesame seed-containing bars (SB or FSB) in this study suggest that it may be one of those four compounds. Whether SES and its metabolites that are excreted in bile undergo enterohepatic circulation and are partly metabolized by the bacterial flora to ED and EL (Fig. 1) remains to be established. The specific conversion of SMN and sesaminol to ED and EL is also yet to be determined.

When sesame seed was combined with flaxseed, the conversion of SECO and other precursors in flaxseed to ED and EL was lower than what was expected from the conversion seen with sesame seed or flaxseed alone. The urinary mammalian lignan recovery relative to the plant lignans was also lower in the FSB group than what was expected from the recoveries in the FB and SB groups. These suggest that the conversion of precursors such as SECO in flaxseed may have been suppressed in the presence of sesame lignans, but the reason for this is currently unknown. Nonetheless, the combined flaxseed and sesame seed appear to still produce mammalian lignans that did not differ significantly from that of flaxseed or sesame seed alone.

In conclusion, precursors in whole unground flaxseed and sesame seed in food bar formulation can be converted to mammalian lignans in postmenopausal women. These food bars may be used to study the anticancer effects and other health benefits of flaxseed and sesame seed, alone and in combination. Because the mammalian lignan production with sesame seed, alone or in combination with flaxseed, was equivalent to that from flaxseed, sesame seed may be used as an alternative to flaxseed as a very rich source of mammalian lignan precursors. The large amount of mammalian lignans produced with sesame seed, reported for the first time here, suggests that they should also be examined for their potential role on some of the reported beneficial effects of sesame seed and its major lignans.

Acknowledgments and Notes

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