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Glucosinolate Composition of Young Shoots and Flower Buds of Capers (*Capparis* Species) Growing Wild in Turkey

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The content and glucosinolate composition of young shoots and raw flower buds of *Capparis spinosa* var. *spinosa* and *Capparis ovata* Desf. var. *canescens* at three different sizes ($x \leq 8$ mm, $8 < x \leq 13$ mm, and $x > 13$ mm) were investigated by HPLC with UV detection. Samples were harvested in August 2001 in Turkey. Twelve different glucosinolates were identified in the young shoots and buds of both species. Total content of glucosinolates ranged from 6.55 μmol/g (large buds of *C. spinosa*) to 45.56 μmol/g (young shoots of *C. ovata*). The main glucosinolate was glucocapperin, which amounted to ~90% of the total glucosinolates. In both species the total glucosinolate content varied in dependence on the bud size, whereas a greater variability was given for buds from *C. spinosa*.

KEYWORDS: Capers; Capparaceae; *Capparis spinosa*; *Capparis ovata*; glucosinolates; HPLC

INTRODUCTION

Capers ("gevil, gebere, gebreotu, kapari, kebere" in Turkish) are the flower buds of *Capparis* genus plants, which are members of the family Capparaceae. These plants are widespread in tropical or subtropical as well as arid areas of the world, and capers have been used for several purposes since ancient times (*1*, *2*).

One group of bioactive components occurring in capers are glucosinolates. Glucosinolates are natural substances, found in many plants, but mainly in the family Crucifere. Many plants of this family are used in agriculture and nutrition, for example, rapeseed, wintercress, false flax, crambe, Brussels sprouts, radish cabbage, broccoli, or cauliflower (*3*−*6*). More than 100 different glucosinolates are known (*7*). Glucosinolates are relatively nontoxic (*8*), but they gain importance from the fact that the products of a myrosinase [thioglucoside glucohydrolase (EC 3.2.3.1)]-induced degradation adversely affect animal growth, reproduction, and performance as well as intake and palatability of fodder. Degradation products also cause goiter and abnormalities in the internal organs of animals (*9*, *10*). On the other hand, it is known that glucosinolates are responsible for the anticarcinogenic activity of *Brassica* vegetables (*11*).

Glucosinolates can also be found in members of Capparaceae. Some published data refer to specific aspects of the qualitative composition of flavonoids, the occurrence of elemental sulfur, and physical and chemical properties (*1*, *2*, *12*−*14*), but up to now there is little information available regarding the glucosinolate content and composition of members of this family. Different glucosinolates such as neoglucobrassicin, 4-methoxyglucobrassicinglucocapperin, glucoiberin, sinigrin, 1-methoxy-3-indolylymethyl, and glucobrassicin were described (*3*, *15*−*18*), but most investigations dealt with roots or leaves.

There are no detailed studies of the glucosinolate content and distribution in young shoots and raw flower buds of capers at different stages of maturity and size. The aim of this investigation was to identify the primary glucosinolates of buds and shoots and to determine if the size of the flower bud affected composition. Two different species of *Capparis* grown in the wild in Turkey were examined.

MATERIALS AND METHODS

Plant Material and Chemicals. Young shoots and flower buds of wild-growing plants of *Capparis spinosa* var. *spinosa* and *Capparis ovata* Desf. var. *canescens* (Coss.) Heywood, respectively, were collected from the south (*C. spinosa*) and the middle (*C. ovata*) of Turkey in August 2001. Raw buds were classified into three different sizes: $x \leq 8$ mm, $8 < x \leq 13$ mm, and $x > 13$ mm. The samples were put into paper bags and then stored over 3 h in an ice box during the transport to the laboratory. Directly afterward, the plant materials were dried to amounts between 11 and 13% moisture in the air without direct sunlight. Colored glass vessels were used for samples at refrigerator temperature (8 °C). Samples were stored under these conditions for about a month before analysis of the glucosinolates.

Extraction of Desulfoglucosinolates. Desulfoglucosinolates were determined according to a modified method described by Fiebig and Jörden (*19*). In brief, 200 mg of the sample material was extracted twice with 70% (v/v) hot methanol at 75 °C for 10 min by ultrasonic treatment after the addition of the internal standard glucotropaeolin (5 and 20 mmol, respectively). The moisture of the shoots was 11%, whereas that of the buds was 13%. These contents were taken into account for the calculation of the individual glucosinolates. Then 2 mL of the crude extract was added on a strong anion-exchange column [SAX 500 mg (Merck, Darmstadt, Germany)] for solid phase extraction,

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Table 1. Mean Glucosinolate Composition of Different Parts of Two *Capparis* Species (Micromoles per Gram of Wet Weight Plant Material)[a]

		C. spinosa var. *spinosa*				*C. ovata* Desf. var. *canescens*			
trivial name	radical name	young shoot	small buds $x \leq 8$ mm	medium buds $8 < x > 13$ mm	large buds $x \geq 13$ mm	young shoot	small buds $x \leq 8$ mm	medium buds $8 < x > 13$ mm	large buds $x \geq 13$ mm
glucocapperin	methyl	35.44 ± 1.12	14.07 ± 0.92	10.75 ± 2.87	5.41 ± 1.62	46.52 ± 3.54	24.23 ± 2.89	22.08 ± 3.54	24.17± 2.89
glucoiberin	3-methylsulfinylpropyl	0.27 ± 0.11	0.15 ± 0.13	0.19 ± 0.15	0.10 ± 0.08	0.11 ± 0.10	0.12 ± 0.11	0.53 ± 0.11	0.16 ± 0.12
progoitrin	(2*R*)-2-hydroxybut-3-enyl	0.10 ± 0.04	0.19 ± 0.09	0.21 ± 0.18	0.15 ± 0.09	0.21 ± 0.11	0.13 ± 0.10	0.44 ± 0.18	0.34 ± 0.21
epiprogoitrin	(2*S*)- 2-hydroxybut-3-enyl	0.26 ± 0.09	0.22 ± 0.20	0.22 ± 0.21	0.12 ± 0.11	0.10 ± 0.09	0.13 ± 0.09	1.44 ± 0.41	0.39 ± 0.11
sinigrin	allyl	0.17 ± 0.15	0.07 ± 0.04	0.27 ± 0.12	0.20 ± 0.10	0.04 ± 0.02	0.03 ± 0.01	0.00 ± 0.01	0.24 ± 0.14
gluconapoleiferin	(2*R*)-2-hydroxypent-4-enyl	0.14 ± 0.11	0.13 ± 0.11	0.46 ± 0.24	0.32 ± 0.21	0.08 ± 0.06	0.42 ± 0.12	0.80 ± 0.21	0.53 ± 0.17
glucoalyssin	5-methylsulfinylpentyl	0.45 ± 0.08	0.46 ± 0.21	0.41 ± 0.27	0.10 ± 0.09	0.42 ± 0.15	0.03 ± 0.02	0.16 ± 0.14	1.05 ± 0.21
gluconapin	but-3-enyl	0.33 ± 0.15	0.12 ± 0.11	0.25 ± 0.09	0.13 ± 0.11	0.14 ± 0.10	0.08 ± 0.02	0.26 ± 0.15	0.28 ± 0.09
4-hydroxyglucobrassicin	4-hydroxyindol-3-ylmethyl	2.04 ± 0.53	0.01 ± 0.01	0.07 ± 0.04	0.02 ± 0.01	0.01 ± 0.01	0.01 ± 0.01	0.03 ± 0.02	0.03 ± 0.02
glucobrassicanapin	pent-4-enyl	0.36 ± 0.12	0.08 ± 0.05	0.27 ± 0.12	0.06 ± 0.02	0.10 ± 0.09	0.06 ± 0.02	0.15 ± 0.02	0.12 ± 0.11
glucobrassicin	indol-3-ylmethyl	0.46 ± 0.23	0.05 ± 0.04	0.08 ± 0.05	0.02 ± 0.01	0.03 ± 0.02	0.03 ± 0.03	0.10 ± 0.09	0.07 ± 0.05
gluconasturtiin	phenethyl	0.22 ± 0.19	0.24 ± 0.18	0.14 ± 0.12	0.13 ± 0.11	0.13 ± 0.09	0.25 ± 0.05	0.36 ± 0.12	0.41 ± 0.14
total content		**40.25**	**15.80**	**13.29**	**6.76**	**47.90**	**25.52**	**26.34**	**27.80**

[a] Values presented as mean values of five replications ± standard deviation.

Figure 1. HPLC chromatogram of the desulfoglucosinolates extracted from young shoots of *C. spinosa*.

which was conditioned with 2 mL of 70% methanol. Unwanted compounds were washed from the column with 2 mL of bidistilled water, and then the pH value of the column was adjusted by using 1 mL of sodium acetate buffer (pH 4). Afterward, the glucosinolates on the column were treated with the enzyme sulfatase (1 mL of a solution of 10 mg of sulfatase/25 mL of water) overnight, which leads to the formation of desulfoglucosinolates. The neutral desulfoglucosinolates were eluted from the column using 1 mL of bidistilled water, whereas all non-glucosinolate anions remained on the exchange column. The solution obtained was used for the HPLC.

The sample preparation was done for each sample five times, and the mean values were calculated. A statistical examination was carried out by calculating the standard deviation.

HPLC. The HPLC analysis was conducted using a Merck-Hitachi low-pressure gradient system, fitted with a L-7100 pump, a Merck-Hitachi L-4250 UV−vis detector set at 229 nm, and a Knauer ChromGate for Windows integration system. Forty microliters of the desulfoglucosinolate-containing eluates was injected by an AS-4000 autosampler onto a 250 × 4 mm, 5 μm LiChrospher 100 RP-18e column (Merck) used with a flow rate of 1 mL/min. The mobile phase used consisted of water (A) versus acetonitrile (B) for a total running time of 43 min, and the gradient changed as follows: in 2.5 min from 100% A to 95% A/5% B, then in 18 min to 80% A/20% B; after a 5 min isocratic period, in 1.5 min back to 100% A. The column was equilibrated at 100% A for 16 min.

Calculation of Each Glucosinolate. For the correct identification of most of the peaks in the chromatogram a reference standard (BCR RM367) was run with the samples. Additionally, seeds of *Iberis amara* and *Lesquerella fendleri* were used for the identification and determination of glucoiberin, which is the main glucosinolate of this plant. Glucocapparin was identified by the preparation of the standard substance. The calculation of each glucosinolate identified in the samples was done by evaluation of the chromatograms obtained by UV detection at 229 nm as described in the EC standard method (*20*). The moisture of the plant material was determined gravimetrically, and the results were taken into account for the calculation of the glucosinolates. The content of each glucosinolate was calculated and expressed as micromoles per gram of plant material. Statistical parameters, such as precision, repeatability, and reproducibility, for the calculation of each glucosinolate were given in the EC standard method.

Statistical Analysis. Data were analyzed by analysis of standard deviation. Student's *t* test to evaluate the statistical significance for independent and variables interactions was performed with two-tailed *t* tests at $P = 0.005$. The data were evaluated using a computer program (Statgraphics).

RESULTS AND DISCUSSION

In all, 12 different glucosinolates were identified in the young shoots and buds of both species (**Table 1**). An example of an HPLC chromatogram of the separation of desulfoglucosinolates of young shoots of *C. spinosa* is given (**Figure 1**). The total

content of glucosinolates ranged from 6.76 μmol/g in large buds of *C. spinosa* to 40.25 μmol/g in young shoots of *C. ovata*. Young shoots of both species had the highest contents of glucosinolates, whereas the contents of the buds were significantly lower ($P < 0.005$). The contents of glucosinolates in the buds of different sizes varied in both species. In *C. spinosa*, glucosinolate content in the buds decreased as size increased. Although the differences between small and medium buds of this species were not significant ($P < 0.005$), there was a significant difference ($P < 0.005$) in the total glucosinolates of small and medium buds, on the one hand, and large buds, on the other. For the total glucosinolates of buds from *C. ovata* a slight increase could be determined as the buds grew larger, but this increase was not significant ($P < 0.005$).

In comparison with other glucosinolate-containing plants or seeds, the amounts found in buds of two different species of *Capparis* were comparable with results found in Brussels sprouts (25.1 μmol/g of dry mass) (*21*) or in seeds of *L. fendleri*, an oilseed used as a renewable resource (27.5 μmol/g of seed material) (*22*). Other widely consumed cruciferous vegetables, such as cabbage, cualiflower, or broccoli, showed lower amounts in total glucosinolates than the investigated buds (*21*, *23*). The amounts of glucosinolates found in shoots were much higher than in other normally consumed vegetables (*21*); only in oilseeds used as renewable resources [*B. verna* (71 μmol/g of seed material), *B. vulgaris* (91.3 μmol/g of seed material), or *L. campestre* (200.9 μmol/g of seed material)] could higher contents of total glucosinolates be found (*22*).

From the composition of the glucosinolates it was conspicuous that glucocapperin was the main glucosinolate of shoots and buds. This result agreed with the investigation of Ahmed et al. (*17*), who found glucocapperin as the main glucosinolate in plants of *Capparis* spp. Also, Kjaer and Thomsen (*15*) found aliphatic glucosinolates to be predominant in leaf and shoot material of the genus *Capparis*, whereas indole glucosinolates occurred only in trace amounts.

In our investigation glucocapperin ranged from 80% of total glucosinolates in large buds of *C. spinosa* to 97% in shoots of *C. ovata*. Most of the values for percent of total glucosinolates varied between 84 and 89%. The only other glucosinolates detected in appreciable amounts were 4-hydroxyglucobrassicin, an indole glucosinolate in the shoots of *C. spinosa* (2.04 μmol/g), and epiprogoitrin in medium buds of *C. ovata* (1.44 μmol/g). All other glucosinolates were found in amounts of <0.8 μmol/g. Schraudolf (*18*) also found only trace amounts of these glucosinolates, but in roots and leafs. Also, in most of the normally consumed crucifereous vegetables and glucosinolate-containing oilseeds, only one main glucosinolate could be found (*21*, *22*).

LITERATURE CITED

(1) Rodrigo, M.; Lazaro, M. J.; Alvarruiz, A.; Giner, V. Composition of capers (*Capparis spinosa*): Influence of cultivar, size and harvest date. *J. Food Sci.* **1992**, *57*, 1152−1154.

(2) Özcan, M.; Akgül, A. Influence of species, harvest date and size on composition of capers (*Capparis* spp.) flower buds. *Nahrung* **1998**, *42*, 102−105.

(3) Daxenbichler, M. E.; Spencer, G. F.; Carlson, D. G.; Rose, G. B.; Brinker, A. M.; Powel, R. G. Glucosinolate composition of seeds from 297 species of wild plants. *Phytochemistry* **1991**, *30*, 2623−2638.

(4) Shahidi, F.; Daun, J. K.; DeClercq, D. R. Glucosinolates in *Brassica* Oilseeds: Processing effects and extraction. In *Antinutrients and Phytochemicals in Food*; Shahidi, F., Ed.; American Chemical Society: Washington, DC, 1995; pp 152−170.

(5) Jezek, J.; Haggett, B. G. D.; Atkinson, A.; Rawson, D. M. Determination of glucosinolates using their alkaline degradation and reaction with ferricyanide. *J. Agric. Food Chem.* **1999**, *47*, 4669−4674.

(6) Matthäus, B.; Luftmann, H. Glucosinolates in members of the family Brassicaceae: separation and identification by LC/ESI-MS-MS. *J. Agric. Food Chem.* **2000**, *48*, 2234−2239.

(7) Kjaer, A.; Skrydstrup, T. Selenoglucosinolates: Synthesis and Enzymatic Hydrolysis. *Acta Chem. Scand.* **1987**, *41*, 29−33.

(8) Bell, M. Nutrients and toxicants in rapeseed meal: a review. *J. Anim. Sci.* **1984**, *58*, 996−1010.

(9) Mawson, R.; Heaney, R. K.; Zdunczyk, Z.; Kozlowska, H. Rapeseed meal-glucosinolates and their antinutritional effects. Part 4. Goitrogenicity and internal organs abnormalities in animals. *Nahrung* **1994**, *38*, 178−191.

(10) Mawson, R.; Heaney, R. K.; Zdunczyk, Z.; Kozlowska, H. Rapeseed meal-glucosinolates and their antinutritional effects. Part 5. Animal reproduction. *Nahrung* **1994**, *38*, 588−598.

(11) Mithen, R. F.; Dekker, M.; Verkerk, R.; Rabot, S.; Johnson, I. T. The nutritional significance, biosynthesis and bioavailability of glucosinolates in human food. *J. Sci. Food Agric.* **2000**, *80*, 967−984.

(12) Brevard, H.; Brambilla, M.; Chaintreau, A.; Marion, J. P. Occurrence of elemental sulphur in capers (*Capparis spinosa* L.) and first investigation of the flavour profile. *Flavour Fragrance J.* **1992**, *7*, 313−321.

(13) Özcan, M.; Akgül, A. Physical and chemical properties of pickled capers (*Capparis* spp.) flower buds. *Obst-, Gemüse- Kartoffelverarbeitung* **2000**, *85*, 165−167.

(14) Inocencio, C.; Rivera, D.; Alcaraz, F.; Tomas-Barberan, F. A. Flavonoid content of commercial capers (*Capparis spinosa*, *C. sicula*, and *C. orientalis*) produced in Mediterranean countries. *Eur. Food Res. Technol.* **2000**, *212*, 70−74.

(15) Kjaer, A.; Thomsen, H. Isothiocyanate-producing glucosides in species of Capparidaceae. *Phytochemistry* **1963**, *2*, 29−32.

(16) Hu, Z.; Lewis, J. A.; Hanley, B.; Fenwick, G. R. 2-Hydroxymethyl glucosinolate from *Capparis masaikai* of Chineese origin. *Phytochemistry* **1989**, *24*, 1252−1254.

(17) Ahmed, Z. F.; Rizk, A. M.; Hammouda, F. M.; Sheif El-Nasr, M. M. Gluosinolates of Egyptian *Capparis* species. *Phytochemistry* **1972**, *11*, 251−256.

(18) Schraudolf, H. Indole glucosinolates of *Capparis spinosa*. *Phytochemistry* **1989**, *88*, 259−260.

(19) Fiebig, H.-J.; Jörden, M. HPLC of desulfoglucosinolates. *J. High-Resolut. Chromatogr. Chromatogr. Commun.* **1990**, *13*, 377−379.

(20) EC Determination of the oilseed glucosinolate content by HPLC. *J. Eur. Communities* **1990**, *L170*, 03.07.27−34.

(21) Kushad, M. M.; Brown, A. F.; Kurilich, A. C.; Juvik, J. A.; Klein, B. P.; Wallig, M. A.; Jeffery, E. H. Variation of glucosinolates in vegetable crops of *Brassica oleacea*. *J. Agric. Food Chem.* **1999**, *47*, 1541−1548.

(22) Matthäus, B.; Luftmann, H. Glucosinolates in members of the family Brassicaceae: Separation and identification by LC/ESI-MS-MS. *J. Agric. Food Chem.* **2000**, *48*, 2234−2239.

(23) Carlson, D. G.; Daxenbichler, M. E.; VanEtten, C. H.; Kwolek, W. F.; Williams, P. H. Glucosinolates in crucifer vegetables: broccoli, Brussels sprouts, cauliflower, collards, kale, mustard greens, and kohlrabi. *J. Am. Soc. Hortic. Sci.* **1987**, *112*, 173−178.

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