

# Dietary Intake of Individual Glucosinolates in Participants of the EPIC-Heidelberg Cohort Study

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dietary intake of individual glucosinolates. The database can be used for epidemiological research on the role of glucosinolates in health and disease.

## Abstract

**Background/Aims:** To compile a database on content of individual glucosinolates in food and to describe the dietary intake of individual glucosinolates in a German population.

**Methods:** Studies analysing the content of individual glucosinolates in food were aggregated to form a database of 26 individual glucosinolates in 18 vegetables and condiments consumed in Germany. This database was linked to food intake data derived from 24-hour diet recalls of 2,121 participants of the EPIC-Heidelberg cohort study. **Results:** Mean total glucosinolate intake ( $\pm$  standard error) was 14.2 ( $\pm$ 1.1) mg/day for men and 14.8 ( $\pm$ 1.3) mg/day for women. The intake increased with age and education; smokers ingested less glucosinolates than never or former smokers. The quantitatively most important individual glucosinolates were glucobrassicin and sinigrin with mean daily intakes of 3.5 ( $\pm$ 0.3) and 1.7 ( $\pm$ 0.2) mg/day for men, and 4.2 ( $\pm$ 0.4) and 2.5 ( $\pm$ 0.4) mg/day for women, respectively. Broccoli, Brussels sprouts and cauliflower contributed most to the total glucosinolate intake in this population. **Conclusions:** The established database allowed for the first time the estimation of

## Introduction

Glucosinolates are sulphur-containing plant metabolites commonly found in cruciferous vegetables. Due to the structural diversity of the side chain about 120 different compounds can be distinguished, but only a small subset of glucosinolates is present in foods commonly consumed by humans. Upon cell rupture, glucosinolates are cleaved by the plant enzyme myrosinase and, depending on the side chain structure, isothiocyanates or indole metabolites can be formed. These breakdown products are responsible for the typical pungent flavour associated with *Brassica* vegetables and are considered as part of the plant defence system [1, 2].

Evidence from cell and animal experiments indicates that these compounds might exert positive health effects [3]. Especially the anticarcinogenic properties of glucosinolate breakdown products are currently under investigation, since isothiocyanates and indoles were shown to be potent modulators of the detoxification system (detoxifying enzymes) [4].

**Table 1.** Individual glucosinolates in food [16, 23, 24, 30–72]

	Cauli- flower	Broc- coli	Red cabbage	Savoy cabbage	White cabbage	Sauer- kraut	Brussels sprouts	Kohl- rabi	Kale	Chin. cabbage	Turnip	Swede
Glucoibervirin	1.60	0.00	0.50	0.36	0.86	0.27	0.01	1.80	0.23	–	0.76	–
Glucoerucin	0.15	0.13	0.83	0.00	0.00	–	0.00	6.46	0.00	–	2.95	6.58
Dehydroerucin	0.00	0.00	0.00	0.00	0.00	–	0.00	0.00	0.00	0.00	0.00	0.00
Glucoberteroin	–	–	–	–	–	–	–	–	–	–	3.92	11.09
Glucoiberin	3.28	1.59	5.01	19.56	7.85	0.82	10.26	1.80	14.68	0.00	0.00	0.00
Glucoaphanin	0.20	22.21	9.10	0.54	0.17	0.45	1.97	0.44	0.37	0.05	0.07	3.14
Glucoaphenin	0.41	0.00	0.00	0.00	0.00	–	0.00	0.82	0.00	–	0.00	–
Glucoalyssin	0.00	0.88	0.61	0.90	0.00	–	0.68	0.38	0.00	0.00	1.30	1.87
Glucocheirolin	0.00	0.00	–	–	0.00	–	2.76	0.00	0.00	–	0.00	–
Glucoerysolin	–	–	1.13	2.49	1.02	–	–	–	–	–	0.00	0.00
Glucoapparin	–	0.00	–	–	–	–	0.00	–	–	–	–	0.32
Glucochlearin	–	–	–	–	–	–	–	–	–	–	1.31	6.76
Sinigrin	4.74	0.19	3.77	17.06	16.31	0.49	44.50	0.30	12.47	0.00	0.60	0.00
Gluconapin	0.10	0.22	2.80	1.06	0.66	0.07	3.56	0.00	0.04	0.00	15.50	1.56
Gluco brassicanapin	0.00	1.08	0.00	0.00	0.00	–	0.00	0.00	0.00	4.30	9.30	2.03
Progoitrin	0.86	0.70	3.47	1.23	1.76	0.04	14.02	0.06	0.82	1.67	13.00	36.18
Epiprogoitrin	0.00	0.16	–	–	–	–	0.00	–	0.00	–	–	–
Napoleiferin	0.00	0.61	0.00	0.00	0.00	–	0.97	0.00	0.00	1.70	2.40	3.07
Gluco tropaeolin	0.00	0.00	0.02	0.00	0.00	–	0.00	0.00	0.00	–	0.00	–
Gluconasturtiin	0.00	0.37	0.15	0.00	0.18	0.00	0.68	0.36	0.17	1.60	16.80	10.52
Sinabin	–	–	–	–	–	–	–	–	–	–	–	–
Gluco barbarin	–	0.31	–	–	–	–	–	–	–	–	–	–
Gluco brassicin	19.01	16.95	7.53	20.55	13.60	6.96	43.82	4.90	15.11	2.93	1.33	4.70
4-Hydroxygluco brassicin	0.56	1.93	1.44	0.76	0.68	–	4.11	0.75	0.65	0.05	2.43	0.94
Neogluco brassicin	2.65	7.91	0.05	0.54	0.54	–	0.48	2.01	1.30	0.73	2.32	7.25
4-Methoxygluco brassicin	1.36	2.07	2.24	3.83	2.79	–	5.53	0.25	3.35	2.88	0.37	1.54
Sum of GLS	34.92	57.31	38.64	68.89	46.42	9.10	133.33	20.32	49.20	15.91	74.36	97.56

Data are medians, presented in milligrams per 100 g fresh weight. GLS = Glucosinolates.

Epidemiological work on the association of dietary glucosinolates and cancer is currently hampered due to the lack of a suitable food composition database for glucosinolates which is the prerequisite to estimate glucosinolate intake. McNaughton and Marks [5] published the first database on the content of total glucosinolates in different vegetables in 2003. Due to structural differences in glucosinolates and subsequently in glucosinolate breakdown products, potentially different biological effects of the single glucosinolates may exist which is also supported by cell studies [4]. This argues for quantification of the intake of individual glucosinolates.

Therefore, the aim of this study was to establish a database on the individual glucosinolate content of foods commonly consumed. Subsequently, these data are used to evaluate the intake of individual glucosinolates in a German population.

## Subjects and Methods

### Database Compilation

A literature search was conducted to identify studies analysing the glucosinolate content of foods commonly consumed by humans. The online databases Medline, BIOSIS Previews, Web of Science (from 1996 onward) and AGRICOLA were accessed during November 2006 and April 2007 to scan for articles containing the term 'glucosinolate'. Additional studies were found by sorting through the reference list of previously identified articles. Likewise, citations in review papers were considered.

Studies eligible for inclusion in the database had to meet the following criteria. The studies had to provide quantitative data on individual glucosinolates (or isothiocyanates) measured by high-performance liquid chromatography (HPLC) or gas chromatography (GC). The analysed foods should have been cultivated for human consumption and no special cultivation treatment should have been applied. Furthermore, studies had to be published in English or German. Of the 117 studies identified, 46 met all inclusion criteria and were used to compile a database on the content of 26 individual glucosinolates in 18 different vegetables or condiments. To allow comparability between these studies, the gluco-

Radish	Horse-radish	Mus-tard	Capers	Water cress	Garden cress
0.00	-	-	-	-	-
0.75	-	-	-	3.26	-
49.54	-	-	-	-	-
-	-	-	-	-	-
0.00	-	-	-	0.00	-
0.64	-	-	-	0.00	-
2.16	-	-	-	-	-
0.00	-	-	-	0.00	-
0.00	-	-	-	-	-
-	-	-	-	-	-
-	-	-	11.33	-	-
-	-	-	-	-	-
0.00	67.56	29.83	-	0.00	-
0.00	-	-	-	0.84	-
0.00	-	-	-	0.00	-
0.16	-	-	-	0.00	-
-	-	-	-	-	-
0.00	-	-	-	0.00	-
0.00	-	-	-	-	1,069.22
0.00	46.58	-	-	133.50	0.00
-	-	29.83	-	14.30	-
-	-	-	-	-	-
0.87	-	-	-	52.24	-
0.60	-	-	-	0.00	-
0.00	-	-	-	0.00	-
1.64	-	-	-	32.25	-
56.35	114.14	59.65	11.33	236.39	1,069.22

sinolate content was converted to milligram/100 g fresh weight based on molecular weights of individual glucosinolates and fresh weight value of the respective vegetables as reported in the literature [6]. Data on isothiocyanates were converted to the corresponding glucosinolate value, assuming that 1 mol of the isothiocyanate corresponds to 1 mol of the according glucosinolate. In case of studies reporting several values for the same food, for example, for different varieties or different harvesting dates, data were used to calculate a mean value for each individual glucosinolate. After compilation of all data from the selected studies, median values for the content of individual glucosinolate for the 18 foods were computed and included in the final database (table 1).

#### Study Population

The database was used to calculate the intake of individual glucosinolates in a subgroup of the European Prospective Investigation into Cancer and Nutrition (EPIC)-Heidelberg cohort, for which 24-hour diet recalls were available. The EPIC-Heidelberg cohort contributes to the EPIC, a multicenter prospective study primarily established to explore the relation between diet, various lifestyle factors and chronic disease risk [7]. Between 1994 and 1998, 25,540 inhabitants of Heidelberg and surrounding commu-

nities aged 35–65 years in females and 40–65 years in males completed a baseline examination, during which data on diet, lifestyle and anthropometry were collected [8]. Dietary information was assessed by a semi-quantitative self-administered food frequency questionnaire. In order to compare dietary data obtained by different food frequency questionnaires used across EPIC-Europe, a second dietary assessment tool, standardized computerised 24-hour recalls by means of the EPIC-SOFT software, was applied to a representative subsample of the EPIC cohorts [9]. In Heidelberg, 2,121 participants completed the 24-hour diet recall between June 1996 and October 1998; the participation rate was 91%. These 2,121 participants (1,034 men and 1,087 women) comprise the study population for the current analysis. All participants gave informed consent at recruitment. This study was approved by the ethical committee of Heidelberg medical school.

#### Data Collection

In a random subset of the cohort, single 24-hour diet recalls per participant were collected on all days of the week (according to a random list), although Fridays were finally under-represented due to a lower availability of the participants for conducting the recall on the following Saturday [10]. Dietary data were assembled through trained interviewers using the software EPIC-SOFT. A detailed description of EPIC-SOFT can be found elsewhere [11]. In brief, a detailed description and quantification of foods (including beverages), recipes and supplements consumed on the recalled day was obtained from each participant. Mixed recipes were broken down into ingredients, thus allowing the consideration of glucosinolate-containing foods used as ingredients for intake calculations. The reported foods were retrieved from a food database implemented in the software and automatically coded. Quantification was done in standard units, standard portions or household measures with the additional help of a photo book showing different foods in different portion sizes. Further information about the cooking method used or the edible part consumed was considered for intake calculation.

Food intake data was linked to the German Food Code and Nutrient Data Base BLS II.3 for calculation of energy and nutrient intake [12, 13]. Furthermore, the food intake was linked with the newly compiled database on glucosinolate content of different foods and condiments which allowed intake calculation of 26 different glucosinolates for each participant.

Lifestyle and sociodemographic data were obtained via self-administered questionnaires and computer-guided face-to-face interviews during baseline examination [14]. Age was measured continuously and grouped in 10-year age classes (35–45, 46–55 and 56–65). Following standardized procedures, weight and height were measured during the participant's visit at the study centre at baseline examination [15]. Body mass index (BMI) was calculated as body weight in kilograms divided by the square of body height in meters and BMI groups (<25, 25–30 and ≥30 kg/m<sup>2</sup>) were created. For participants with missing data on height (n = 3) and weight (n = 1), the sex-specific median was imputed and those with missing information on educational attainment (n = 2) were categorized into the none or primary group.

#### Statistical Analysis

Descriptive statistics of the study population are given as percentages for categorical variables and mean and standard deviation for continuous variables. Single 24-hour recalls were used to

**Table 2.** Variability of analytical data on sinigrin and glucobrassicin content in selected foods

	Sinigrin			Glucobrassicin		
	median	Q1–Q3	min–max	median	Q1–Q3	min–max
Cauliflower	4.7	3.4–14.2	0.7–30.1	19.0	7.1–26.4	2.8–127.8
Broccoli	0.2	0–0.3	0–0.5	17.0	8.5–32.8	4.3–190.6
Red cabbage	3.8	2.7–9.3	1.1–11.9	7.5	7.1–15.3	3.9–19.6
Savoy cabbage	17.1	15.5–17.9	5.1–37.0	20.6	14.1–34.3	10.8–59.8
White cabbage	16.3	10.3–21.7	3.6–43.3	13.6	6.9–16.9	2.1–22.1
Sauerkraut	0.5	–	–	7.0	–	–
Brussels sprouts	44.5	23.7–47.7	3.1–55.7	43.8	27.1–62.4	12.0–267.2
Kohlrabi	0.3	0–2.5	0–8.0	4.9	3.9–5.0	3.0–5.1
Kale	12.5	9.7–22.8	1.7–52.7	15.1	7.6–30.9	2.2–67.8
Chinese cabbage	0	–	–	2.9	2.7–3.0	2.4–3.0
Turnip	0.6	0.3–1.2	0–1.8	1.3	0.7–2.6	0–3.8
Swede	0	0–0.2	0–0.3	4.7	4.5–6.3	4.3–7.8
Radish	0	0–0	0–0	0.9	0.6–1.1	0.6–2
Horseradish	67.6	–	–	–	–	–
Mustard	29.8	–	–	–	–	–
Watercress	0	–	–	52.2	–	–

Data are presented in milligrams per 100 g fresh weight. Capers and garden cress were omitted since they contain neither sinigrin nor glucobrassicin. Q1–Q3 = Interquartile range; min–max = minimum to maximum; – = not enough values/studies available to determine interquartile range/minimum and maximum.

describe dietary intake at the group level. Accordingly, average daily intake of glucosinolates in milligram/day is given for men and women and by age groups, presented as mean and standard error of mean. To compare the intake of glucosinolates in different subgroups according to lifestyle and sociodemographic factors, Wilcoxon and Kruskal-Wallis tests were used with a significance level of  $\alpha = 0.05$ . The contribution of food sources to the average glucosinolate intake is expressed as mean percentage of total intake. All statistical analyses were performed with SAS 9.1 (SAS Institute Inc., Cary, N.C., USA).

## Results

The content of individual glucosinolates in different vegetables included in the database is shown in table 1. Some glucosinolates like glucobrassicin or sinigrin are present in nearly all listed foods, while others are limited to few specific plants. For example, noteworthy amounts of glucocapparin can be found exclusively in capers and glucotropaeolin is a clear indicator for garden cress. To give an impression of the variability of glucosinolate contents for different foods in our database, we showed the interquartile range and the minimum and maximum values of the glucosinolates sinigrin and glucobrassicin in table 2.

Characteristics of the study population for which the glucosinolate intake was calculated are given in table 3. The mean intake ( $\pm$  standard error) of total glucosinolates was  $14.2 \pm 1.1$  mg/day for men and  $14.8 \pm 1.3$  mg/day for women (table 4) in the EPIC-Heidelberg cohort study. Men ingested 6.6 mg/1,000 kcal total glucosinolates, which is distinctly lower than 9.1 mg/1,000 kcal calculated for women. More than half of the ingested glucosinolates had an aliphatic side chain structure (7.8 mg/day for men and 8.4 mg/day for women), and sinigrin, dehydroerucin, glucoraphanin and glucoiberin were quantitatively the most important aliphatic glucosinolates. With 5.0 mg/day in men and 5.7 mg/day in women, indolylglucosinolates contributed 35 and 38%, respectively, to the total glucosinolate intake. The intake of glucobrassicin as the main representative of this group was 3.5 mg/day in men and 4.2 mg/day in women. The lowest contribution to overall glucosinolate intake was provided by the group of aromatic glucosinolates with an average contribution of 10% in men and 5% in women. Total glucosinolate intake in men and women did not significantly differ. However, in men and women, the mean intake rose with higher age ( $p = 0.004$ ) (table 5). Similarly, glucosinolate intake increased significantly with educational attainment ( $p = 0.03$ ) and smokers ingested less total

**Table 3.** Description of the EPIC-Heidelberg study population participating in 24-hour diet recalls (1996–1998)

	Men (n = 1,034)		Women (n = 1,087)	
	n	%	n	%
Age, years (mean ± SD)	1,034	53.7 ± 7.0	1,087	50.3 ± 8.5
Height, cm (mean ± SD)	1,034	175.6 ± 6.5	1,087	164.0 ± 6.2
Weight, kg (mean ± SD)	1,034	84.0 ± 12.6	1,087	68.4 ± 12.5
BMI, kg/m <sup>2</sup> (mean ± SD)	1,034	27.2 ± 3.8	1,087	25.5 ± 4.8
Energy intake, kcal/day (mean ± SD)	1,034	2,429.5 ± 873.8	1,087	1,823.7 ± 627.2
Age categories				
≤45 years	139	13	349	32
45–55 years	406	39	337	31
>55 years	489	47	401	37
BMI categories (kg/m <sup>2</sup> )				
<25	304	29	586	54
25–30	512	50	323	30
≥30	218	21	178	16
Educational attainment				
None or primary	349	34	285	26
Secondary/technical	362	35	554	51
Longer education	323	31	248	23
Smoking status				
Never	358	35	559	51
Former	446	43	308	28
Current	230	22	220	20

glucosinolates than never or former smokers ( $p = 0.01$ ). BMI was not associated with intake. Similar results were obtained when intake of glucosinolates was expressed per 1,000 kcal or intake of glucosinolate subgroups was evaluated.

Vegetables contributing most to the intake of total glucosinolates were broccoli, Brussels sprouts and cauliflower (table 6). This is also true for the subgroup of aliphatic glucosinolates and indolylglucosinolates. Aromatic glucosinolates occur only in few vegetables in noteworthy amounts. Most of them were coded as recipe components like mustard, cress or capers which contributed most to the intake of this subgroup.

## Discussion

So far, most epidemiological studies on the association between glucosinolates or glucosinolate breakdown products and disease risk used food consumption data, that is, intake of cruciferous vegetables, as exposure measure. This might be due to the lack of appropriate glucosinolate databases. This study is the first to establish a database

on glucosinolate content of foods consumed by humans which focuses not just on the total amount of glucosinolates but on the individual compounds. This distinction seems reasonable since glucosinolates form a variety of different breakdown products depending on the side chain structure, which in turn might lead to divergent biological effects. For example, sulforaphane, the breakdown product of glucoraphanin, is a potent inducer of phase II enzymes in cultured human cells. Glucobrassicin is metabolized to indole-3-carbinol and the subsequently formed acidic condensation products have been shown to influence also the activity of phase I enzymes [4].

In 2003, McNaughton and Marks [5] summarized the available literature consisting of 18 studies to form the first database on total glucosinolate contents of cruciferous vegetables; all of these studies which reported the amount of individual glucosinolates are included in our database. Additionally, further studies were identified by extensive literature search and carefully assessed before entering them into the database. It was our basic aim to establish a database which can be used to estimate the intake of individual glucosinolates in a German popula-

**Table 4.** Mean ( $\pm$  standard error of the mean) intake of total glucosinolates, glucosinolate subgroups and individual glucosinolates in men and women of the EPIC-Heidelberg study who provided a 24-hour diet recall (n = 2,121)

	Men	Women
Total glucosinolates	14.20 $\pm$ 1.12	14.80 $\pm$ 1.29
Aliphatic glucosinolates		
Glucoibervirin	0.20 $\pm$ 0.02	0.19 $\pm$ 0.02
Glucoerucin	0.30 $\pm$ 0.05	0.22 $\pm$ 0.03
Dehydroerucin	1.69 $\pm$ 0.29	1.60 $\pm$ 0.22
Glucoberberoin	0.05 $\pm$ 0.02	0.03 $\pm$ 0.01
Glucoiberin	1.01 $\pm$ 0.10	1.14 $\pm$ 0.13
Glucoraphanin	1.48 $\pm$ 0.21	1.23 $\pm$ 0.18
Glucoraphenin	0.12 $\pm$ 0.01	0.11 $\pm$ 0.01
Glucoalyssin	0.10 $\pm$ 0.01	0.09 $\pm$ 0.01
Glucocheirolin	0.01 $\pm$ 0.01	0.07 $\pm$ 0.02
Glucoerysolin	0.09 $\pm$ 0.01	0.07 $\pm$ 0.01
Glucocapparin	0.02 $\pm$ 0.00	0.02 $\pm$ 0.00
Glucocochlearin	0.02 $\pm$ 0.01	0.01 $\pm$ 0.00
Sinigrin	1.69 $\pm$ 0.19	2.46 $\pm$ 0.37
Gluconapin	0.29 $\pm$ 0.05	0.26 $\pm$ 0.04
Glucobrassicinapin	0.17 $\pm$ 0.03	0.13 $\pm$ 0.02
Progoitrin	0.48 $\pm$ 0.07	0.67 $\pm$ 0.11
Epiprogoitrin	0.01 $\pm$ 0.00	0.01 $\pm$ 0.00
Napoleiferin	0.07 $\pm$ 0.01	0.08 $\pm$ 0.01
Total	7.81 $\pm$ 0.59	8.37 $\pm$ 0.75
Indolylglucosinolates		
Glucobrassicin	3.54 $\pm$ 0.33	4.15 $\pm$ 0.42
Hydroxyglucobrassicin	0.28 $\pm$ 0.03	0.32 $\pm$ 0.04
Neoglucobrassicin	0.68 $\pm$ 0.08	0.60 $\pm$ 0.07
Methoxyglucobrassicin	0.50 $\pm$ 0.04	0.58 $\pm$ 0.06
Total	5.01 $\pm$ 0.45	5.65 $\pm$ 0.55
Aromatic glucosinolates		
Glucotropaeolin	0.60 $\pm$ 0.52	0.30 $\pm$ 0.20
Gluconasturtiin	0.42 $\pm$ 0.08	0.31 $\pm$ 0.06
Sinialbin	0.36 $\pm$ 0.05	0.17 $\pm$ 0.02
Glucobarbarin	0.02 $\pm$ 0.00	0.01 $\pm$ 0.00
Total	1.39 $\pm$ 0.53	0.79 $\pm$ 0.21

Data are presented in milligrams per day.

tion. Thus, studies reporting glucosinolate data on seeds, seedlings or parts of the plants normally not consumed by humans were discarded. Similarly, varieties grown as fodder or of no marketable quality were not included. If a special cultivation method was used, only the results for the control group were taken. Whenever indicated that a certain variety was cultivated for the European market, only those values were entered into the database. This is the case especially for radish, since the varieties for Eu-

rope are cultured to have less pungent flavour and, therefore, less dehydroerucin which is the main glucosinolate in radish [16]. Finally, the focus was on vegetables consumed by participants of the EPIC-Heidelberg study; thus, the glucosinolate-containing vegetables rocket, rape, wasabi, papaya or some Asian cabbage varieties like bok choy, kai lan or kai choi were not included here. Concerning the analytical methods for food analysis, only results from GC- or HPLC-based methods for glucosinolate determination were entered into the database, since comparability of both methods in terms of precision can be assumed [17]. Other methods to analyse glucosinolate or respective isothiocyanate levels in food are the glucose release method [18] and the cyclocondensation assay [19]. However, both methods quantify only the total amount and do not measure individual glucosinolates.

For most vegetables or condiments, several studies were available; thus, an average value (median) for the glucosinolate contents was calculated. Nevertheless, for some foods the data basis was quite scarce. As a result, the values for horseradish, capers, mustard and sauerkraut are all based each on the result of 1 study. The reported values of glucosinolate contents of a certain food varied considerably, which led to the decision to calculate median instead of mean values. The high variability is easily explained by natural variation, since a lot of factors influence the amount of glucosinolates in a plant. Besides the cultivar, harvest time and growing conditions, like soil type, water supply or fertilisation, affect the glucosinolate content [20]. However, small differences in analytical methods or during sample preparation and handling can impact on the results of the measurement and therefore contribute to the variability.

For all above-mentioned reasons, glucosinolate intake estimates based on our glucosinolate database will differ from the true intake of an individual. Still, true intake values can only be determined by means of the duplicate method, that is, chemical analysis of the consumed diet, an approach which is simply not feasible in epidemiological settings with large sample sizes or with a focus on usual food consumption over a prolonged time period.

Linking this database with food consumption data of participants of the EPIC-Heidelberg cohort who completed a 24-hour diet recall yielded a mean intake of total glucosinolates of 14.2 mg/day for men and 14.8 mg/day for women. In comparison, in the Spanish EPIC cohort a mean intake of total glucosinolates of 6.8 mg/day for men and 6.2 mg/day for women was found [21]. These intake estimates were computed based on dietary data obtained via a diet history method (assessing habitual food intake)

**Table 5.** Mean ( $\pm$  standard error of the mean) intake of glucosinolates and glucosinolate subgroups in men and women of the EPIC-Heidelberg study who provided a 24-hour diet recall (n = 2,121), by age groups

	Men			Women		
	$\leq 45$ years	45–55 years	$> 55$ years	$\leq 45$ years	45–55 years	$> 55$ years
Total glucosinolates	11.00 $\pm$ 2.35	12.70 $\pm$ 1.68	16.40 $\pm$ 1.80	11.70 $\pm$ 1.82	15.40 $\pm$ 2.40	17.00 $\pm$ 2.40
Aliphatic glucosinolates	7.10 $\pm$ 1.64	6.93 $\pm$ 0.92	8.76 $\pm$ 0.87	6.68 $\pm$ 1.04	8.09 $\pm$ 1.34	10.10 $\pm$ 1.34
Indolyl glucosinolates	3.39 $\pm$ 0.87	4.92 $\pm$ 0.81	5.53 $\pm$ 0.62	4.64 $\pm$ 0.81	5.86 $\pm$ 1.01	6.36 $\pm$ 1.01
Aromatic glucosinolates	0.50 $\pm$ 0.10	0.88 $\pm$ 0.21	2.07 $\pm$ 1.11	0.39 $\pm$ 0.06	1.49 $\pm$ 0.66	0.55 $\pm$ 0.66

Data are presented in milligrams per day.

and linked with the database on total glucosinolate content of food compiled by McNaughton and Marks [5]. The higher intake of glucosinolates in EPIC-Heidelberg should be mainly due to a higher consumption of cabbages in EPIC-Heidelberg [22] and the consideration of more glucosinolate-containing vegetables for intake calculation in our study. Furthermore, the intake estimates in EPIC-Spain accounted for losses of glucosinolates through food storage and processing. However, results are also affected by differences in the dietary assessment methods and the applied glucosinolate database.

Some studies reporting glucosinolate intake estimates performed their calculations using glucosinolate data obtained by their own laboratory analyses of food. Hrnčirik and Velisek [23] reported a mean intake of total glucosinolates of 4.7 mg/day for the Czech Republic by linking the result of HPLC-based glucosinolate analyses of 5 different *Brassica* vegetables to the aggregated Czech food consumption data from 1993. Other studies reported distinctly higher mean intake estimates for total glucosinolates. A study from the United Kingdom used food intake data of the 1980 National Food Survey to estimate a mean total glucosinolate intake of 29.4 mg/day from cooked and 46.1 mg/day from fresh vegetables [24]. Another German study reported a mean intake of total glucosinolates of 43.1 mg/day per person [25] based on the per capita consumption (from trade statistics) of cabbage, cauliflower and Brussels sprouts for the year 1989. In a following publication, the same author confirmed his earlier results by estimating the glucosinolate intake of a representative sample of the region Potsdam for 1992/93 to be 41.1 mg/day [26]. Thus, in comparison to our estimates, these latter intake values are considerably higher, which is even more surprising, since their intake estimates are based only on 10 glucosinolate-containing vegetables in contrast to the 18 vegetables we used. However, in the

**Table 6.** Percent contribution of selected vegetables to the dietary intake of total, aliphatic, indolyl and aromatic glucosinolates in participants of the EPIC-Heidelberg study who provided a 24-hour diet recall (n = 2,121)

	Glucosinolates, % of total intake			
	total	aliphatic	indolyl	aromatic
Broccoli	19	16	26	3
Brussels sprouts	14	15	15	1
Cauliflower	13	8	25	0
Radish	13	22	2	0
White cabbage	9	10	9	0
Red cabbage	6	7	4	0
Savoy cabbage	5	6	5	0
Swede/turnip	4	5	1	12
Mustard	3	3	0	22
Miscellaneous <sup>1</sup>	8	4	1	57

<sup>1</sup> Recipe components like mustard, cress, capers or horseradish.

Potsdam study the glucosinolate content of vegetables was obtained from the researchers' own analyses with the glucose-release method which yielded generally much higher values than the GC- or HPLC-based results included in our database. Additionally, differences in food choices between the Potsdam and the Heidelberg study populations might help to explain the observed differences in glucosinolate intakes, since a higher cabbage intake in the Potsdam study [27] eventually will lead to higher glucosinolate intake values.

Next to the differences in dietary assessment methods applied and differences in glucosinolate level assigned to foods, several other factors could affect estimates of glucosinolate intake and, thus, complicate direct compari-

son of study results. We included the consumption of some glucosinolate-containing foods like mustard, turnip or radish in our intake calculation, but these foods/condiments were often not considered in other studies. Furthermore, dietary habits differ not only between countries but might also vary according to regional habits or time within the same country. Additionally, some population characteristics like the age structure, educational attainment or smoking status impact on food consumption and therefore on glucosinolate intake, as shown in the present study or in the Spanish EPIC cohort [21]. These effects may be more pronounced if studies are small and not population based. Furthermore, storage time, processing conditions and preparation methods have an effect on glucosinolate content of foods that is not negligible [28]. For example, cooking leads to an average loss of glucosinolates of 36% in cruciferous vegetables [24]. However, the decrease depends on the food analysed, ranging from 28% for cabbage to 48% for swede/turnip. Similarly, the decrease is not uniform over the individual glucosinolates, ranging from 34% for sinigrin to 59% for neoglucobrassicin [24]. Since the methods applied for food preparation are manifold, and numerous factors influencing the food content of glucosinolates during processing still need to be evaluated, we refrain from reporting glucosinolate losses due to preparation method or considering them in our intake analyses. Thus, the glucosinolate intake data presented here represent an overestimation of the true intake.

The strength of our results is the use of a database specifically designed to evaluate the intake of individual glucosinolates taking most exact food analysis data from all relevant foods providing glucosinolates into account.

Furthermore, dietary data is obtained at an individual level via a standardized computer-assisted interview. These data can be used to estimate average intake of glucosinolates at the group level as done in the current study, since a sufficiently large number of subjects was interviewed [9]. Participants of the EPIC-Heidelberg cohort study show more favourable socioeconomic and health-related indicators than the underlying population [29]. Thus, the estimates of glucosinolate intake presented here are not fully representative of the intake of the underlying population.

In conclusion, we could establish a database on the food contents of individual glucosinolates for a variety of vegetables and condiments. The linkage with dietary data of participants of the EPIC-Heidelberg cohort yielded total glucosinolate intake values within the range of other published studies that used databases on total glucosinolate content of foods. Furthermore, this study is the first to report intake data on individual glucosinolates. The results presented here form a basis for future epidemiological research into possible associations between intake of glucosinolates and occurrence of diseases such as cancer where biological mechanisms are already hypothesised.

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### References

- 1 Mithen RF: Glucosinolates and their degradation products. *Adv Bot Res* 2001;35:213–262.
- 2 Fahey JW, Zalcmann AT, Talalay P: The chemical diversity and distribution of glucosinolates and isothiocyanates among plants. *Phytochemistry* 2001;56:5–51.
- 3 International Agency for Research on Cancer: IARC Handbooks of Cancer Prevention, vol 9: Cruciferous vegetables, Isothiocyanates and Indoles. Lyon, IARC Press, 2008.
- 4 Higdon JV, Delage B, Williams DE, Dashwood RH: Cruciferous vegetables and human cancer risk: epidemiologic evidence and mechanistic basis. *Pharmacol Res* 2007; 55:224–236.
- 5 McNaughton SA, Marks GC: Development of a food composition database for the estimation of dietary intakes of glucosinolates, the biologically active constituents of cruciferous vegetables. *Br J Nutr* 2003;90:687–697.
- 6 Souci SW, Fachmann W, Kraut H: Food Composition and Nutrition Tables, ed 6. Stuttgart, Medpharm, 2000.
- 7 Riboli E, Kaaks R: The EPIC Project: rationale and study design. *European Prospective Investigation into Cancer and Nutrition. Int J Epidemiol* 1997;26(suppl 1):S6–S14.
- 8 Boeing H, Wahrendorf J, Becker N: EPIC-Germany – a source for studies into diet and risk of chronic diseases. *European Investigation into Cancer and Nutrition. Ann Nutr Metab* 1999;43:195–204.
- 9 Kaaks R, Riboli E: Validation and calibration of dietary intake measurements in the EPIC project: methodological considerations. *European Prospective Investigation into Cancer and Nutrition. Int J Epidemiol* 1997; 26(suppl 1):S15–S25.
- 10 Slimani N, Kaaks R, Ferrari P, Casagrande C, Clavel-Chapelon F, Lotze G, Kroke A, Trichopoulos D, Trichopoulou A, Lauria C, Bellegotti M, Ocke MC, Peeters PHM, Engset D, Lund E, Agudo A, Larranaga N, Mattisson I, Andren C, Johansson I, Davey G, Welch AA, Overvad K, Tjonneland A, Van Staveren WA, Saracci R, Riboli E: European



- Prospective Investigation into Cancer and Nutrition (EPIC) calibration study: rationale, design and population characteristics. *Public Health Nutr* 2002;5:1125–1145.
- 11 Slimani N, Deharveng G, Charrondiere RU, van Kappel AL, Ocke MC, Welch A, Lagiou A, van LM, Agudo A, Pala V, Brandstetter B, Andren C, Stripp C, Van Staveren WA, Riboli E: Structure of the standardized computerized 24-h diet recall interview used as reference method in the 22 centers participating in the EPIC project. *European Prospective Investigation into Cancer and Nutrition. Comput Methods Programs Biomed* 1999;58:251–266.
  - 12 Voss S, Charrondiere UR, Slimani N, Kroke A, Riboli E, Wahrendorf J, Boeing H: EPIC-SOFT a European computer program for 24-hour dietary protocols. *Z Ernährungswiss* 1998;37:227–233.
  - 13 Bundesinstitut für gesundheitlichen Verbraucherschutz und Veterinärmedizin (BgVV): Der Bundeslebensmittelschlüssel (BLS II.3): Konzeption, Aufbau und Dokumentation der Datenbank blsd. Berlin, BgVV-Hefte, 1999.
  - 14 Kroke A, Bergmann MM, Lotze G, Jeckel A, Klipstein-Grobusch K, Boeing H: Measures of quality control in the German component of the EPIC study. *European Prospective Investigation into Cancer and Nutrition. Ann Nutr Metab* 1999;43:216–224.
  - 15 Klipstein-Grobusch K, Georg T, Boeing H: Interviewer variability in anthropometric measurements and estimates of body composition. *Int J Epidemiol* 1997;26(suppl 1):S174–S180.
  - 16 Carlson DG, Daxenbichler ME, VanEtten CH, Hill CB, Williams PH: Glucosinolates in radish cultivars. *J Amer Soc Hort Sci* 1985;110:634–638.
  - 17 Hrnčirik K, Velisek J, Davidek J: Comparison of HPLC and GLC methodologies for determination of glucosinolates using reference material. *Z Lebensm Unters Forsch A* 1998;206:103–107.
  - 18 Heaney RK, Spinks EA, Fenwick GR: Improved method for the determination of the total glucosinolate content of rapeseed by determination of enzymically released glucose. *Analyst* 1988;113:1515–1518.
  - 19 Zhang YS, Wade KL, Prestera T, Talalay P: Quantitative determination of isothiocyanates, dithiocarbamates, carbon disulfide, and related thiocarbonyl compounds by cyclocondensation with 1,2-benzenedithiol. *Anal Biochem* 1996;239:160–167.
  - 20 Rosa EAS, Heaney RK, Fenwick GR, Portas CAM: Glucosinolates in crop plants. *Hortic Rev* 1997;19:99–215.
  - 21 Agudo A, Ibanez R, Amiano P, Ardanaz E, Barricarte A, Berenguer A, Dolores CM, Dorransoro M, Jakszyn P, Larranaga N, Martinez C, Navarro C, Pera G, Quiros JR, Sanchez MJ, Tormo MJ, Gonzalez CA: Consumption of cruciferous vegetables and glucosinolates in a Spanish adult population. *Eur J Clin Nutr* 2008;62:324–331.
  - 22 Agudo A, Slimani N, Ocke MC, Naska A, Miller AB, Kroke A, Bamia C, Karalis D, Vineis P, Palli D, Bueno-de-Mesquita HB, Peeters PH, Engeset D, Hjartaker A, Navarro C, Martinez GC, Wallstrom P, Zhang JX, Welch AA, Spencer E, Stripp C, Overvad K, Clavel-Chapelon F, Casagrande C, Riboli E: Consumption of vegetables, fruit and other plant foods in the European Prospective Investigation into Cancer and Nutrition (EPIC) cohorts from 10 European countries. *Public Health Nutr* 2002;5:1179–1196.
  - 23 Hrnčirik K, Velisek J: Glucosinolate content of common *Brassicaceae* family vegetables. *Potrav Vedy* 1997;15:161–172.
  - 24 Sones K, Heaney RK, Fenwick GR: An estimate of the mean daily intake of glucosinolates from cruciferous vegetables in the UK. *J Sci Food Agric* 1984;35:712–720.
  - 25 Lange R, Baumgrass R, Diedrich M, Henschel KP, Kujawa M: Glucosinolate in der Ernährung – Pro und Contra einer Naturstoffklasse. *Ernährungs-Umschau* 1992;39:252–257.
  - 26 Lange R, Lushtak B, Ozierenski B, Pfaff G: Glucosinolate in Kohlgemüse: Natürliche Schadstoffe oder protektive Faktoren?; in *Neue Aspekte der gesundheitlichen Wirkung pflanzlicher Lebensmittel*. Quedlinburg, DGQ, 1994, vol 29, pp 49–73.
  - 27 Schulze MB, Brandstetter BR, Kroke A, Wahrendorf J, Boeing H: Quantitative food intake in the EPIC-Germany cohorts. *European Investigation into Cancer and Nutrition. Ann Nutr Metab* 1999;43:235–245.
  - 28 de Vos RH, Blijleven WGH: The effect of processing conditions on glucosinolates in cruciferous vegetables. *Z Lebensm Unters Forsch* 1988;187:525–529.
  - 29 Boeing H, Korfmann A, Bergmann MM: Recruitment procedures of EPIC-Germany. *European Investigation into Cancer and Nutrition. Ann Nutr Metab* 1999;43:205–215.
  - 30 Anderson JL, Lisk DJ, Stoewsand GS: Glucosinolates in rutabaga grown in soil capped over coal fly ash. *J Food Sci* 1990;55:556–557.
  - 31 Betz JM, Fox WD: High-performance liquid chromatographic determination of glucosinolates in Brassica vegetables; in Huang MT, Osawa T, Ho CT (eds): *Food Phytochemicals for Cancer Prevention I*. Washington DC, American Chemical Society, 1994, pp 181–196.
  - 32 Carlson DG, Daxenbichler ME, VanEtten CH: Glucosinolates in crucifer vegetables – turnips and rutabagas. *J Agric Food Chem* 1981;29:1235–1239.
  - 33 Carlson DG, Daxenbichler ME, VanEtten CH: Glucosinolates in crucifer vegetables – broccoli, Brussels-sprouts, cauliflower, collards, kale, mustard greens, and kohlrabi. *J Am Soc Hort Sci* 1987;112:173–178.
  - 34 Carlson DG, Daxenbichler ME, Tookey HL: Glucosinolates in turnip tops and roots – cultivars grown for greens and/or roots. *J Am Soc Hort Sci* 1987;112:179–183.
  - 35 Chong C, Ju HY, Bible BB: Glucosinolate composition of turnip and rutabaga cultivars. *Can J Plant Sci* 1982;62:533–536.
  - 36 Chung FL, Morse MA, Eklind KI: Quantitation of human uptake of the anticarcinogen phenethyl isothiocyanate after a watercress meal. *Cancer Epidemiol Biomarkers Prev* 1992;1:383–388.
  - 37 Ciska E, Kozłowska H: The effect of cooking on the glucosinolates content in white cabbage. *Eur Food Res Technol* 2001;212:582–587.
  - 38 Ciska E, Pathak DR: Glucosinolate derivatives in stored fermented cabbage. *J Agric Food Chem* 2004;52:7938–7943.
  - 39 Ciska E, Martyniak-Przybyszewska B, Kozłowska H: Content of glucosinolates in cruciferous vegetables grown at the same site for two years under different climatic conditions. *J Agric Food Chem* 2000;48:2862–2867.
  - 40 Ciska E, Piskula M, Martyniak-Przybyszewska B: Glucosinolates in various cabbage cultivars grown in Poland. *Pol J Food Nutr Sci* 1994;3:119–126.
  - 41 Gil V, MacLeod AJ: Benzylglucosinolate degradation in *Lepidium sativum*: effects of plant age and time of autolysis. *Phytochem* 1980;19:1365–1368.
  - 42 Gliszczynska-Swiglo A, Ciska E, Pawlak-Lemanska K: Changes in the content of health-promoting compounds and antioxidant activity of broccoli after domestic processing. *Food Addit Contamin* 2006;23:1088–1098.
  - 43 Goodrich RM, Parker RS, Lisk DJ: Glucosinolate, carotene and cadmium content of *Brassica oleracea* grown on municipal sewage sludge. *Food Chemistry* 1988;27:141–150.
  - 44 Heaney RK, Fenwick GR: Glucosinolates in *Brassica* vegetables – analysis of 22 varieties of Brussels sprout (*Brassica oleracea* var. gemmifera). *J Sci Food Agric* 1980;31:785–793.
  - 45 Hecht SS, Carmella SG, Kenney PMJ: Effects of cruciferous vegetable consumption on urinary metabolites of the tobacco-specific lung carcinogen 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone in Singapore Chinese. *Cancer Epidemiol Biomarkers Prev* 2004;13:997–1004.
  - 46 Kang JY, Khalid EI, Juvik JA: Genetic and environmental effects on glucosinolate content in Chinese cabbage. *Hortscience* 2006;41:1382–1385.

- 47 Kushad MM, Brown AF, Kurilich AC: Variation of glucosinolates in vegetable crops of *Brassica oleracea*. J Agric Food Chem 1999; 47:1541–1548.
- 48 Lewis J, Fenwick GR: Glucosinolate content of *Brassica* vegetables Chinese cabbages pe-tsai (*Brassica pekinensis*) and pak-choi (*Brassica chinensis*). J Sci Food Agric 1988; 45:379–386.
- 49 Lockwood GB, Belkhir A: Glucosinolate spectrum of some Algerian *Cruciferae*. Pl Syst Evol 1991;176:11–20.
- 50 Nilsson J, Olsson K, Engqvist G: Variation in the content of glucosinolates, hydroxycinnamic acids, carotenoids, total antioxidant capacity and low-molecular-weight carbohydrates in *Brassica* vegetables. J Sci Food Agric 2006;86:528–538.
- 51 Rodrigues AS, Rosa EAS: Effect of post-harvest treatments on the level of glucosinolates in broccoli. J Sci Food Agric 1999;79:1028–1032.
- 52 Rosa EAS, Heaney RK: The effect of cooking and processing on the glucosinolate content – studies on 4 varieties of Portuguese cabbage and hybrid white cabbage. J Sci Food Agric 1993;62:259–265.
- 53 Rosa EAS, Rodrigues AS: Total and individual glucosinolate content in 11 broccoli cultivars grown in early and late seasons. Hortscience 2001;36:56–59.
- 54 Schonhof I, Krumbein A, Brückner B: Genotypic effects on glucosinolates and sensory properties of broccoli and cauliflower. Nahrung/Food 2004;48:25–33.
- 55 Shattuck VI, Kakuda Y, Shelp BJ: Chemical composition of turnip roots stored or intermittently grown at low temperature. J Am Soc Hort Sci 1991;116:818–822.
- 56 Shattuck VI, Kakuda Y, Shelp BJ: Effect of low temperature on the sugar and glucosinolate content of rutabaga. Scientia Horticulturae 1991;48:9–19.
- 57 Sones K, Heaney RK, Fenwick GR: The glucosinolate content of UK vegetables – cabbage, swede, and turnip. Food Addit Contamin 1984;1:289–296.
- 58 Sones K, Heaney RK, Fenwick GR: An estimate of the mean daily intake of glucosinolates from cruciferous vegetables in the UK. J Sci Food Agric 1984;35:762–766.
- 59 Song L, Thornalley PJ: Effect of storage, processing and cooking on glucosinolate content of *Brassica* vegetables. Food Chem Toxicol 2007;45:216–224.
- 60 Tian Q, Rosselot RA, Schwartz SJ: Quantitative determination of intact glucosinolates in broccoli, broccoli sprouts, Brussels sprouts, and cauliflower by high-performance liquid chromatography-electrospray ionization-tandem mass spectrometry. Anal Biochem 2005;343:93–99.
- 61 Tiedink HGM, Davies JAR, van Broekhoven LW: Formation of mutagenic N-nitroso compounds in vegetable extracts upon nitrite treatment: a comparison with the glucosinolate content. Food Chem Toxicol 1988;26: 947–954.
- 62 Tolonen M, Taipale M, Viander B: Plant-derived biomolecules in fermented cabbage. J Agric Food Chem 2002;50:6798–6803.
- 63 Vallejo F, Tomas-Barberan FA, Garcia-Viguera C: Glucosinolates and vitamin C content in edible parts of broccoli florets after domestic cooking. Eur Food Res Technol 2002;215:310–316.
- 64 Vallejo F, Tomas-Barberan FA, Garcia-Viguera C: Health-promoting compounds in broccoli as influenced by refrigerated transport and retail sale period. J Agric Food Chem 2003;51:3029–3034.
- 65 Vallejo F, Garcia-Viguera C, Tomas-Barberan FA: Changes in broccoli (*Brassica oleracea* L. Var. italica) health-promoting compounds with inflorescence development. J Agric Food Chem 2003;51:3776–3782.
- 66 Vallejo F, Tomas-Barberan FA, Garcia-Viguera C: Potential bioactive compounds in health promotion from broccoli cultivars grown in Spain. J Sci Food Agric 2002;82: 1293–1297.
- 67 Vallejo F, Tomas-Barberan FA, Gonzalez Benavente-Garcia A: Total and individual glucosinolate contents in inflorescences of eight broccoli cultivars grown under various climatic and fertilisation conditions. J Sci Food Agric 2003;83:307–313.
- 68 Van Etten CH, Daxenbichler ME, Tookey HL: Glucosinolates – potential toxicants in cabbage cultivars. J Am Soc Hort Sci 1980; 105:710–714.
- 69 Verkerk R, Dekker M: Glucosinolates and myrosinase activity in red cabbage (*Brassica oleracea* L. var. Capitata f. rubra DC.) after various microwave treatments. J Agric Food Chem 2004;52:7318–7323.
- 70 Verkerk R, Dekker M, Jongen WMF: Post-harvest increase of indolyl glucosinolates in response to chopping and storage of *Brassica* vegetables. J Sci Food Agric 2001;81:953–958.
- 71 Vermeulen M, Van den Berg R, Freidig AP: Association between consumption of cruciferous vegetables and condiments and excretion in urine of isothiocyanate mercapturic acids. J Agric Food Chem 2006;54:5350–5358.
- 72 Wennberg M, Ekvall J, Olsson K: Changes in carbohydrate and glucosinolate composition in white cabbage during blanching and treatment with acetic acid. Food Chemistry 2006; 95:226–236.