

## Influence of the Production Method on Phytochemical Concentrations in Whole Wheat (*Triticum aestivum* L.): A Comparative Study

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The present study was performed to evaluate the concentrations of phytochemicals (carotenoids and phenolic acids) in wheat varieties grown under comparable organic and conventional conditions over three years as part of a long-term field trial. Phenolic acids of the hydroxybenzoic acid and hydroxycinnamic acid group were quantified by HPLC-DAD after extraction as free, soluble conjugated, and bound forms. Furthermore, the carotenoids lutein and zeaxanthin were determined by HPLC-DAD. There were no statistically significant differences between the two farming systems (sum of carotenoids ( $\mu\text{g/g}$ ) for 2003,  $0.91 \pm 0.55$  organic vs  $0.96 \pm 0.34$  conventional; for 2005,  $1.61 \pm 0.22$  organic vs  $1.33 \pm 0.19$  conventional; for 2006,  $0.87 \pm 0.33$  organic vs  $0.83 \pm 0.11$  conventional; sum of phenolic acids ( $\mu\text{g/g}$ ) for 2003,  $448.4 \pm 151.1$  organic vs  $327.3 \pm 232.8$  conventional; for 2005,  $502.8 \pm 168.3$  organic vs  $484.4 \pm 111.2$  conventional; for 2006,  $659.1 \pm 112.5$  organic vs  $945.9 \pm 353.6$  conventional). However, statistically significant year-to-year differences up to 55% were observed. Taken together, these results indicate that climate factors have a greater impact on the phytochemical concentrations in whole wheat than the production method (organic/conventional).

**KEYWORDS:** Organic; wheat; carotenoids; phenolic acids

### INTRODUCTION

In Germany the organic agricultural area increased by 30% in the past 10 years (1). Furthermore, consumer demand for organically produced crops is constantly rising (2). This development is part of a change in consumer behavior due to the discussion on safer and healthier food. It has been shown that organically grown fruits and vegetables have lower loads of pesticides than conventionally produced ones (3). Furthermore, it has been discussed if organically grown fruits and vegetables are healthier due to higher concentrations of phytochemicals (e.g., carotenoids and polyphenols) (4, 5).

Lutein and zeaxanthin are the major carotenoids in wheat; lutein accounts for 80–90% of the total carotenoids (6, 7). In different wheat cultivars the concentration of lutein ranges between 26 and 143  $\mu\text{g}/100\text{ g}$  (8). Lutein and zeaxanthin play significant roles in reducing the risk of age-related macular degeneration (9) and cataracts (10). Besides carotenoids, whole wheat contains significant amounts of phenolic acids (e.g., ferulic and vanillic acid). Ferulic acid is the predominant phenolic acid

(up to 90%) with concentrations ranging between 0.5 and 1 mg/g of dry matter (11). Beverages (e.g., coffee, tea, and wine) and cereals are the most important contributors to total polyphenol intake, which is approximately 1 g per day in the Western industrial states (12, 13). The potential health benefits of phenolic acids have been related mostly to their antioxidant capacity (14). Therefore, a regular consumption of whole-grain products rich in carotenoids and polyphenols might result in health benefits.

Several studies have been conducted to gain information on the effect of the production method on the carotenoid and polyphenol concentrations in different fruits and vegetables. Higher carotenoid concentrations were found in organically grown sweet peppers, tomatoes, and yellow plums (15–18). In contrast, lower amounts were reported in organically produced tomatoes (19, 20).

With regard to polyphenols, higher concentrations were found in organically grown apples, strawberry cultivars, and marionberries (21–23). In contrast, no differences in the polyphenol content were observed between organically and conventionally produced yellow plums or strawberries, as well as black currants (17, 24, 25). Other studies demonstrated lower amounts of phenolic compounds in organically grown tomatoes and broccoli (20, 26).

These inconsistencies suggest that factors other than the production method alone might affect the phytochemical concentration, for example, cultivar, microclimate, stage of ripeness,

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and soil conditions. Several of the mentioned studies did not take these factors into account (16, 27, 28). Furthermore, plant foods, for example, whole wheat, have not been studied for differences in the phytochemical concentrations based on the production method. Thus, there is a need for more studies comparing the phytochemical contents in plant foods under well-defined conditions.

Therefore, the aim of the present study was to determine the carotenoid and phenolic acid concentrations in whole wheat grown under identical site conditions in organic and conventional farming systems (DOC trial) in Switzerland over three years (2003, 2005, and 2006).

## MATERIALS AND METHODS

**Chemicals.** Ferulic acid, *p*-hydroxybenzoic acid, caffeic acid, syringic acid, vanillic acid, and lutein as well as zeaxanthin were purchased from Roth (Karlsruhe, Germany). Coumaric acid was obtained from Merck (Darmstadt, Germany) and sinapic acid from Sigma (Taufkirchen, Germany).

All other chemicals were purchased from Sigma-Aldrich or Merck.

**Wheat Samples.** Wheat (*Triticum aestivum* L.) samples were harvested in 2003, 2005, and 2006 from the long-term DOC (dynamic–organic–conventional) field trial near Basel in Switzerland (cultivar Titlis in 2003 and 2005; cultivar Runal in 2006) (29). Materials from two organic farming systems, namely, biodynamic (BIODYN) and bioorganic (BIOORG), and from two conventional systems, one using mineral fertilizer (CONMIN) and the other using mineral fertilizers plus farmyard manure (CONFYM), were used. Additionally, there were plots that received no fertilizer at all (NOFERT) (29). Four samples of each variation, originating from independent field replicate plots of the DOC trial, were used for this study. More details on the farming system and on wheat cultivation of the DOC trial therein are found in Mäder et al. (29, 30). All wheat samples were dried until residual moisture was < 14% and were stored at 4 °C until analysis.

**Carotenoid Extraction from Wheat.** Whole wheat was ground using a laboratory blender (Severin, Sundern, Germany). Half a gram of the whole-wheat flour was further soaked in a saturated NaCl solution for 1 h. Afterward, the whole-wheat flour was homogenized in 50  $\mu$ L of 100 mM EDTA and 5 mL of acetone containing 0.01% BHT using an Ultra Turrax T25 (IKA, Stauffen, Germany). The homogenate was centrifuged at 3000g for 5 min at 4 °C, and the organic extract was collected in a separate tube. Extraction was repeated until the organic extracts were colorless. After the combined organic extracts had been washed with 5 mL of a saturated NaCl solution, the organic solvent was removed using a rotary evaporator (Laborota 4003-digital, Heidolph, Schwabach, Germany). The remaining aqueous phase was extracted three times with 2 mL of *n*-hexane containing 0.01% BHT, and the combined organic extracts were evaporated to dryness under a stream of nitrogen gas. The extraction was repeated five times. The recovery for lutein and zeaxanthin was > 95%. The coefficient of variation of the method was < 5% (intra-assay).

**HPLC Analysis of Carotenoids.** The HPLC analysis was performed as described previously (31). Calibration curves of lutein and zeaxanthin were constructed in the range of 0.025 and 25 mM in which the linearity of the response was given.

**Phenolic Acid Extraction from Wheat.** Whole wheat was milled in a laboratory blender (Severin, Sundern, Germany) to a fine powder as already described for the carotenoids.

The free, soluble conjugated, and bound phenolic acids were extracted from the whole-wheat flour according to previously described methods (32–34) with some modifications.

**Extraction of Free Phenolic Acids.** Half a gram of whole flour was extracted three times with 10 mL of acetone/methanol/water (7:7:6). The mixture was centrifuged, and the supernatants were collected and combined. The organic extract was evaporated under nitrogen gas until only the aqueous phase remained, which was further extracted three times with ethyl acetate. The combined organic extracts were evaporated to dryness under nitrogen gas.

**Extraction of Soluble Conjugated Phenolic Acids.** The aqueous supernatant (see Extraction of Free Phenolic Acids) was treated with 1 mL

of 10 M KOH for 90 min at 37 °C. The resulting hydrolysate was acidified to pH 2 using 6 M HCl and extracted twice with ethyl acetate. The extracts were combined and evaporated to dryness under a gentle stream of nitrogen gas.

**Extraction of Bound Phenolic Acids.** The pellet (see Extraction of Free Phenolic Acids) was treated with 1 mL of 10 M KOH for 90 min at 37 °C, acidified to pH 2 using 6 M HCl, and extracted twice with ethyl acetate. The organic extracts were combined and evaporated to dryness under nitrogen gas.

Free and soluble conjugated phenolic acids were dissolved in 100  $\mu$ L of methanol/water (1:1). The bound phenolic acids were dissolved in 1 mL of methanol/water (1:1). All extractions were repeated five times. The recoveries of the phenolic acids (free, soluble conjugate, and bound phenolic acids) analyses ranged between 72 and 87%. The coefficient of variation of the method was < 10% (intra-assay).

**HPLC Analysis of Phenolic Acids.** HPLC analysis was performed on a high-pressure gradient system from Shimadzu (Duisburg, Germany) equipped with an autoinjector, a photodiode array detector, and a fluorescence detector. Separation was carried out on a Prontosil (150 mm  $\times$  4.6 mm i.d., particle size = 3  $\mu$ m) reversed-phase column (Bischoff, Leonberg, Germany). Solvent A consisted of 0.1% formic acid in water (pH 3) and solvent B of acetonitrile. A linear gradient was used: from 5 to 12% B in 10 min, 12% for 10 min, from 12 to 35% in 20 min, and from 35 to 5% in 1 min. The flow rate was set to 1.2 mL/min, and the injection volume was 70  $\mu$ L for the free phenolic acids, 50  $\mu$ L for the soluble conjugated phenolic acids, and 25  $\mu$ L for the bound phenolic acids. The eluent was recorded with diode array detection at 280 nm for quantification of the hydroxybenzoic acids and at 320 nm of the hydroxycinnamic acids. Peaks were scanned between 190 and 500 nm. Quantification of the polyphenols was performed using commercially available reference compounds as listed above. Calibration curves of the phenolic acids were constructed in the range of 0.1–100  $\mu$ M in which the linearity of the response was given. The quantification of those phenolic acids, which were not commercially available (in this case diferulic acid), was based on a representative standard of the same polyphenol class (in this case ferulic acid). For identification of the phenolic acids, for which no reference compounds were commercially available, a HPLC-MS analysis was performed on a HP 1100 series HPLC (Agilent Technologies, Waldbronn, Germany) equipped with an autoinjector, binary HPLC pump, column heater, UV detector, and HP Chem Station for data collection and handling. The HPLC was interfaced to an HP series 1100 mass selective detector equipped with an atmospheric pressure ionization–electrospray (API-ES) chamber. The wheat polyphenols were analyzed with the following conditions: capillary voltage, 3.5 kV; fragmentor voltage, 150 V; nebulizing pressure, 50 psi; drying gas temperature, 350 °C; drying gas flow, 12.5 L/min. The scan mode was used for data collection. Spectra were scanned over a mass range of *m/z* 100–600 at 0.98 s per cycle. For HPLC the same conditions as described above were used.

**Statistical Analysis.** All statistical calculations were performed using the STATVIEW program version 5.0 (SAS Institute, Cary, NC; 1998). Results are reported as means  $\pm$  SD. Differences between the mean values of carotenoids and phenolic acids in wheat between treatment groups were statistically analyzed using the one-way analysis of variance (ANOVA) and Tukey–Kramer post hoc test. Differences between years were statistically analyzed using the unpaired Student *t* test. Phenolic acid concentrations were transformed logarithmically because the equal variance and normal distribution to allow statistical analysis with ANOVA were rejected. Differences were considered to be significant at *p* < 0.05.

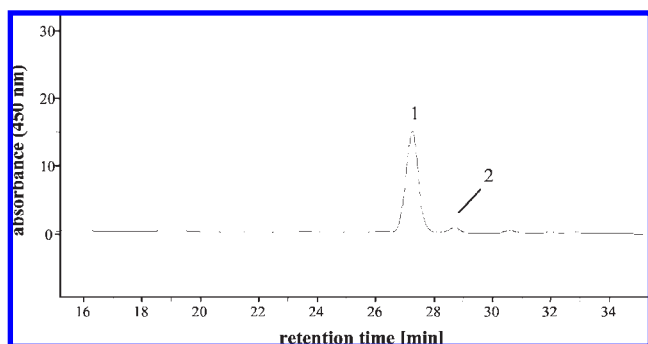
## RESULTS

In this study the carotenoid and polyphenol concentrations in whole wheat were analyzed in one unfertilized (NOFERT) as well as amended with the biodynamic preparations and weeds were mechanically controlled) treatment, in two organic (BIODYN, biodynamic; BIOORG, bioorganic), and in two conventional (CONFYM, mineral fertilizers plus farmyard manure; CONMIN, using mineral fertilizer) farming systems of the DOC trial for a period of 3 years (cultivar Titlis in 2003 as well as in 2005; cultivar Runal in 2006).

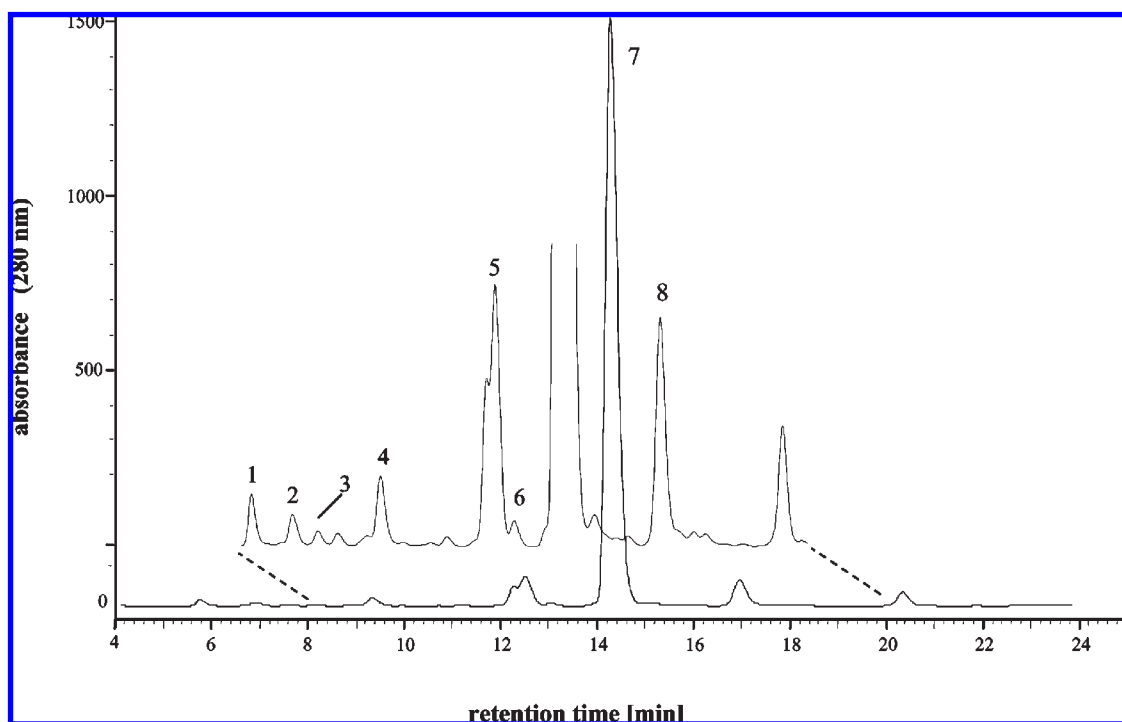
With respect to the carotenoids, we were able to identify lutein and zeaxanthin in whole-grain wheat by comparing their retention times and UV-vis absorbance spectra with those of commercially available reference compounds. The polyphenols identified in the free, soluble conjugated, and bound phenolic acid fractions in wheat using HPLC-MS and reference compounds were as follows: in the group of hydroxybenzoic acids, syringic acid and vanillic acid, as well as *p*-hydroxybenzoic acid; in the group of hydroxycinnamic acids, sinapic acid, ferulic acid, diferulic acid, and coumaric acid, as well as caffeic acid. In **Figures 1** and **2** representative HPLC chromatograms of the phytochemicals are depicted.

**Carotenoids.** *Influence of the Production Method.* In 2003, the CONFYM treatment displayed the highest carotenoid concentration ( $0.97 \pm 0.14 \mu\text{g/g}$ ) compared to the other production methods. No other statistically significant differences between the farming systems were observed (**Table 1**).

In 2005, both organic treatments (BIODYN and BIOORG) had statistically significant higher carotenoid concentrations than the conventional farming systems (CONFYM and CONMIN).



**Figure 1.** Representative HPLC chromatogram of a wheat sample after carotenoid extraction at a wavelength of  $\lambda = 450 \text{ nm}$ : 1, lutein; 2, zeaxanthin.



**Figure 2.** Representative HPLC chromatogram of a wheat sample after phenolic acid extraction of the bound fraction at a wavelength of  $\lambda = 280 \text{ nm}$ : 1, syringic acid; 2, vanillic acid; 3, *p*-hydroxybenzoic acid; 4, caffeic acid; 5, sinapic acid; 6, diferulic acid; 7, ferulic acid; 8, coumaric acid.

However, the untreated treatment (NOFERT) showed statistically significant higher lutein and zeaxanthin contents ( $1.68 \pm 0.14$  and  $0.14 \pm 0.07 \mu\text{g/g}$ , respectively) than the organic and conventional treatments (**Table 1**).

In 2006, the NOFERT treatment exhibited the statistically significant highest lutein and zeaxanthin concentrations ( $1.13 \pm 0.23$  and  $0.15 \pm 0.06 \mu\text{g/g}$ , respectively). Additionally, zeaxanthin concentrations were statistically significant higher in both conventional production methods than in the organic ones (**Table 1**).

*Influence of the Crop Year.* Harvest year 2005 showed statistically significant higher carotenoid concentrations than harvest year 2003 (**Figure 3**).

**Phenolic Acids.** In **Figure 3** and **Table 2** only the sum of the phenolic acids for the three individual fractions (free, soluble conjugated, bound) as well as for all fractions is shown. In the Supporting Information (Tables X–Z) the concentrations of the individual phenolic acids are listed.

**Influence of the Production Method (Free, Soluble Conjugated, Bound Phenolic Acids).** In 2003, in all three fractions the BIODYN ( $594 \pm 57 \mu\text{g/g}$ ) and the NOFERT ( $469 \pm 120 \mu\text{g/g}$ ) treatments showed statistically significant higher phenolic acid contents than the treatments BIODYN, CONFYM, and CONMIN (**Table 2**).

In 2005, the CONMIN treatment exhibited a statistically significant higher phenolic acid content in the fraction of the free, the NOFERT in the fraction of the soluble conjugated, and the BIOORG treatment in the fraction of the bound phenolic acids than the other analyzed treatments. This leads to the statistically significant higher concentration in the sum of all phenolic acids in the BIOORG ( $589 \pm 53 \mu\text{g/g}$ ) treatment than in the BIODYN, CONFYM, and CONMIN treatments (**Table 2**).

In 2006, in all three phenolic acid fractions, the CONFYM treatment showed a statistically significant higher phenolic acid concentration ( $1262 \pm 92 \mu\text{g/g}$ ) compared to the other treatments. No other significant differences were observed (**Table 2**).

**Table 1.** Carotenoid Concentrations of Wheat (*Triticum aestivum* L. Cv. Titlis and Runal) from One Untreated, Two Organic, and Two Conventional Farming Systems<sup>a</sup>

	year	variety	production method				
			BIODYN ( <i>n</i> = 20)	BIOORG ( <i>n</i> = 20)	CONFYM ( <i>n</i> = 20)	CONMIN ( <i>n</i> = 20)	NOFERT ( <i>n</i> = 20)
lutein ( $\mu\text{g/g}$ )	2003	Titlis	0.40 $\pm$ 0.10a	0.77 $\pm$ 0.10a	0.84 $\pm$ 0.13	0.49 $\pm$ 0.11a	0.48 $\pm$ 0.13a
	2005	Titlis	1.40 $\pm$ 0.09ab	1.42 $\pm$ 0.13ab	1.25 $\pm$ 0.21	1.24 $\pm$ 0.16	1.68 $\pm$ 0.14abc
	2006	Runal	0.65 $\pm$ 0.08	0.52 $\pm$ 0.07	0.69 $\pm$ 0.08	0.71 $\pm$ 0.1	1.13 $\pm$ 0.23abc
zeaxanthin ( $\mu\text{g/g}$ )	2003	Titlis	0.08 $\pm$ 0.03a	0.10 $\pm$ 0.02a	0.13 $\pm$ 0.02	0.08 $\pm$ 0.02a	0.08 $\pm$ 0.03a
	2005	Titlis	0.11 $\pm$ 0.05ab	0.12 $\pm$ 0.01ab	0.08 $\pm$ 0.02	0.08 $\pm$ 0.01	0.14 $\pm$ 0.07abc
	2006	Runal	0.10 $\pm$ 0.02ab	0.07 $\pm$ 0.01ab	0.14 $\pm$ 0.01	0.12 $\pm$ 0.03	0.15 $\pm$ 0.06abc
sum of carotenoids ( $\mu\text{g/g}$ )	2003	Titlis	0.47 $\pm$ 0.13a	0.87 $\pm$ 0.12a	0.97 $\pm$ 0.14	0.58 $\pm$ 0.12a	0.56 $\pm$ 0.15a
	2005	Titlis	1.48 $\pm$ 0.12ab	1.54 $\pm$ 0.13ab	1.33 $\pm$ 0.22	1.33 $\pm$ 0.17	1.83 $\pm$ 0.21abc
	2006	Runal	0.74 $\pm$ 0.1	0.60 $\pm$ 0.08	0.83 $\pm$ 0.09	0.83 $\pm$ 0.12	1.28 $\pm$ 0.25abc

<sup>a</sup> NOFERT, no fertilizers; BIODYN, biodynamic; BIOORG, bioorganic; CONFYM, mineral fertilizers plus farmyard manure; CONMIN, using mineral fertilizer. Wheat was harvested in Switzerland in 2003, 2005, and 2006 as part of the DOC long-term field trial. Values are means  $\pm$  SD: a,  $p < 0.05$  vs CONFYM; b,  $p < 0.05$  vs CONMIN; c,  $p < 0.05$  vs BIODYN, BIOORG (ANOVA; Tukey–Kramer post hoc test). Sum of carotenoids is calculated as the sum of the results for the single carotenoids lutein and zeaxanthin.

**Table 2.** Phenolic Acid Concentrations of Wheat (*Triticum aestivum* L. Cv. Titlis and Runal) from One Untreated, Two Organic, and Two Conventional Farming Systems<sup>a</sup>

	year	variety	production method				
			BIODYN ( <i>n</i> = 20)	BIOORG ( <i>n</i> = 20)	CONFYM ( <i>n</i> = 20)	CONMIN ( <i>n</i> = 20)	NOFERT ( <i>n</i> = 20)
sum of free phenolics ( $\mu\text{g/g}$ )	2003	Titlis	4.14 $\pm$ 0.86a	2.92 $\pm$ 0.95	2.95 $\pm$ 1.23	3.24 $\pm$ 1.64	4.08 $\pm$ 1.08a
	2005	Titlis	2.65 $\pm$ 1.30	3.27 $\pm$ 1.67a	2.03 $\pm$ 0.66	3.64 $\pm$ 1.57a	3.13 $\pm$ 1.02a
	2006	Runal	4.39 $\pm$ 1.13a	3.93 $\pm$ 0.49a	14.55 $\pm$ 3.85	7.13 $\pm$ 6.03a	5.13 $\pm$ 1.30a
sum of soluble conjugated phenolics ( $\mu\text{g/g}$ )	2003	Titlis	4.36 $\pm$ 1.45b	2.02 $\pm$ 0.96	4.14 $\pm$ 1.89	1.96 $\pm$ 0.89	11.47 $\pm$ 16.6b
	2005	Titlis	2.24 $\pm$ 1.53	3.62 $\pm$ 1.45c	2.85 $\pm$ 0.51	5.19 $\pm$ 1.99ac	5.99 $\pm$ 4.20ac
	2006	Runal	3.00 $\pm$ 0.72a	3.31 $\pm$ 2.54a	10.33 $\pm$ 2.05	5.69 $\pm$ 2.19a	3.19 $\pm$ 1.18a
sum of bound phenolics ( $\mu\text{g/g}$ )	2003	Titlis	586 $\pm$ 57	278 $\pm$ 31c	326 $\pm$ 325c	315 $\pm$ 82c	454 $\pm$ 114ab
	2005	Titlis	377 $\pm$ 130	529 $\pm$ 54c	451 $\pm$ 89	504 $\pm$ 126c	497 $\pm$ 207
	2006	Runal	631 $\pm$ 117a	663 $\pm$ 87a	1237 $\pm$ 92	617 $\pm$ 182a	631 $\pm$ 117a
sum of phenolic acids ( $\mu\text{g/g}$ )	2003	Titlis	594 $\pm$ 57	283 $\pm$ 31c	333 $\pm$ 325c	320 $\pm$ 84c	469 $\pm$ 120ab
	2005	Titlis	382 $\pm$ 133	589 $\pm$ 53ac	456 $\pm$ 87	513 $\pm$ 126c	506 $\pm$ 207
	2006	Runal	638 $\pm$ 118a	670 $\pm$ 86a	1262 $\pm$ 92	629 $\pm$ 190a	669 $\pm$ 133a

<sup>a</sup> NOFERT, no fertilizers; BIODYN, biodynamic; BIOORG, bioorganic; CONFYM, mineral fertilizers plus farmyard manure; CONMIN, using mineral fertilizer. Wheat was harvested in Switzerland in 2003, 2005, and 2006 as part of the DOK field trial. Values are means  $\pm$  SD: a,  $p < 0.05$  vs CONFYM; b,  $p < 0.05$  vs BIOORG; c,  $p < 0.05$  vs BIODYN; (ANOVA; Tukey–Kramer post hoc test). Sum of phenolic acids is calculated as the sum of the results for the single polyphenols in the three fractions: free, soluble conjugated and bound phenolic acids (sinapic acid, ferulic acid, diferulic acid, coumaric acid, caffeic acid, syringic acid, vanillic acid, *p*-hydroxybenzoic acid).

*Influence of the Crop Year.* Harvest year 2005 showed statistically significant higher phenolic acid concentrations than harvest year 2003 (Figure 3).

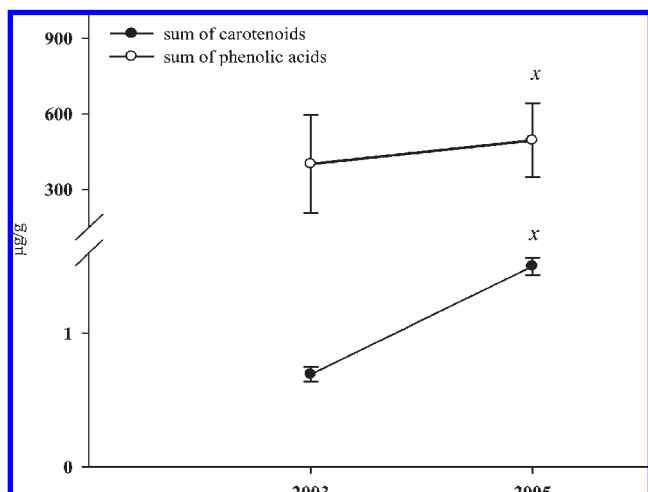
## DISCUSSION

To date, no studies have been published investigating the influence of the production method (organic vs conventional) on the phytochemical concentration in whole wheat over a period of several years. Therefore, the aim of the present study was to evaluate the carotenoid (lutein and zeaxanthin) and phenolic acid concentrations of wheat (cv. Titlis and Runal) grown under well-defined organic and conventional conditions (DOC trial).

The lutein and zeaxanthin concentrations of the whole wheat samples averaged between 0.25 and 1.68  $\mu\text{g/g}$  and between 0.03 and 0.15  $\mu\text{g/g}$ , respectively, with lutein being the major carotenoid (up to 95%). The sum of phenolic acids in whole wheat of the DOC trial ranged between 282 and 1262  $\mu\text{g/g}$ , whereas ferulic acid (approximately 85%) was the predominant phenolic acid. Other important phenolic acids were coumaric, sinapic, and caffeic acid. The majority of the phenolic acids in whole wheat (up to 98%) were bound to cell components. This is well in line with other observations (11, 32, 35, 36). In these studies >97% of the

phenolic acids were bound to cell components. Furthermore, the carotenoid and phenolic acid concentrations in the present study are similar to those previously reported (6, 36–41). For instance, Liyana-Pathirana et al. reported on polyphenol concentrations between 700 and 900  $\mu\text{g/g}$  in wheat grown in Saskatchewan, Canada (41), and Adom et al. reported on lutein concentrations of 0.26–1.43  $\mu\text{g/g}$  in 11 wheat varieties of durum, hard, and soft winter wheat grown in the United States (8).

In our study the organically produced wheat exhibited phytochemical concentrations similar to those of the conventionally grown ones. Other studies compared the phytochemical content of organically and conventionally produced sweet peppers, tomatoes, carrots, celery, and kale as well as apples, strawberry cultivars, and marionberries on the basis of fresh weight. The authors reported higher phytochemical concentrations in the organically produced fruits and vegetables than in the conventionally produced ones (18, 21–23). If phytochemical concentrations are found to be elevated in organically grown fruits and vegetables, an explanation might be that plants change their metabolism toward carbon-containing compounds (starch, cellulose, and non-nitrogen-containing secondary metabolites such as phenolic acids and terpenoids) when nitrogen availability is



**Figure 3.** Influence of the crop year on the carotenoid and phenolic acid concentrations of wheat. Significant differences between the crop year 2003 and 2005 were observed ( $x$ ,  $p < 0.001$  vs 2003, unpaired Students  $t$  test). Sum of carotenoids is calculated as the sum of lutein and zeaxanthin. Sum of phenolic acids is calculated as the sum of the results for the individual polyphenols in the free, soluble, and bound phenolic acid fractions: sinapic acid, ferulic acid, diferulic acid, coumaric acid, caffeic acid, syringic acid, vanillic acid,  $p$ -hydroxybenzoic acid.

limited for growth. Otherwise, when nitrogen is readily available, plants will primarily form compounds with high nitrogen content, for example, proteins for growth and nitrogen-containing secondary metabolites such as alkaloids (21, 42–44). In organic production the nitrogen availability is limited due to a different fertilizing strategy than in the conventional production method (21, 42). Additionally, the accumulation of nitrogen differs between plant organs. In leaves and roots as well as stems the nitrogen accumulation is higher than in fruits and seeds (45). Therefore, different nitrogen fertilizing strategies might have a greater influence on phytochemical concentrations in leaves, roots, and stems than in fruits and seeds. Therefore, it cannot be excluded that differences may occur in other organs. This might explain the missing effects of the production method on phytochemical contents in our study.

In agreement with our observation, no differences in the phytochemical concentrations between the organic and conventional farming systems was reported for yellow plums and strawberries as well as black currants (17, 24, 25). However, comparable studies for wheat do not exist to confirm these results.

Factors such as plant genotype, cultivar, and climate variations have been shown to exhibit greater influence on the phytochemical content in apples and tomatoes than the production method (43, 44, 46). In wheat, significant cultivar differences were also found for carotenoid and phenolic acid concentrations (7, 8, 36, 37, 39). For instance, Adom et al. reported concentrations of ferulic acid between 191 and 303  $\mu\text{mol}/100\text{ g}$  of grain and of lutein between 0.26 and 1.43  $\mu\text{g}/100\text{ g}$  of grain dependent on the wheat variety (8).

We were able to demonstrate year-to-year variations up to 55% for 2003 and 2005, which can be explained by climate differences between the two harvest years. Other studies have also demonstrated that climate factors have a big influence on the phytochemical contents in fruits and vegetables including wheat (7, 27, 28). The lutein content of einkorn cultivar AC Knowles varied statistically significantly over three years, ranging from 6.5  $\mu\text{g}/\text{g}$  (1996) to 9.2  $\mu\text{g}/\text{g}$  (2000) (7).

In conclusion, the organically produced wheat exhibited no statistically significant differences in the phytochemical concen-

trations as compared to the conventionally grown ones. However, it has to be pointed out that climate variations have a greater influence on the phytochemical concentration in whole wheat than the production method.

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**Supporting Information Available:** Tables X, Y, and Z show the concentrations of the individual phenolic acids. This material is available free of charge via the Internet at <http://pubs.acs.org>.

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