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Effects of *Fusarium* infection on the amino acid composition of winter wheat grain

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ABSTRACT

Winter wheat (susceptible cultivar Ritmo) was grown in 2006 near Kiel and in 2007 near Heide in northern Germany. Plants were inoculated at anthesis using a *Fusarium graminearum* macroconidial suspension. The percentage of *Fusarium*-damaged kernels (FDK) ranged from 0 ± 2% to 28 ± 2%. The contents of the *Fusarium* mycotoxin deoxynivalenol (DON) and wheat amino acids were determined in the grain. Levels of the amino acids alanine, lysine, and tyrosine increased with the percentage of FDK or DON contents whereas glutamic acid contents decreased. Aspartic acid and threonine were not related to the percentage of FDK or DON contents. Effects of *Fusarium* infection on other amino acids were significant only at the sampling site with the higher degree of *Fusarium*-damage. Interestingly, those amino acids that increased consistently and significantly with the degree of *Fusarium*-damage are derived from phosphoenolpyruvate or pyruvate, suggesting that pathogen-induced changes in the glycolytic input for amino acid biosynthesis play a significant role for the amino acid composition of *Fusarium*-damaged winter wheat grain. On average, amino acid contents decreased by 0.13% compared to the amino acid content of sound kernels upon an increase of 1% of FDK.

1. Introduction

The genus *Fusarium* includes fungi with saprophytic as well as plant-pathogenic capabilities. Wheat plants are infected at anthesis, via the open flowers, when humidity is sufficiently high to allow fungal growth (Beyer, Röding, Ludewig, & Verreet, 2004; Beyer, Verreet, & Ragab, 2005; Beyer, Klix, Klink, & Verreet, 2006). *Fusarium* species produce mycotoxins with adverse effects on the physiology of other organisms, including humans (Bondy & Pestka, 2000; Speijers & Speijers, 2004). *Fusarium* mycotoxins belonging to the group of trichothecenes inhibit eukaryotic protein biosynthesis (Rotter, Prelusky, & Pestka, 1996). Amino acids are the basic structural building units of peptides and proteins. The total storage protein content was lower in *Fusarium*-damaged kernels than in sound kernels but no significant qualitative differences in protein were detected by various analytical methods (Nightingale, Marchylo, Clear, Dexter, & Preston, 1999). The amino acid composition of *Fusarium*-damaged soybean did not vary between infected and sound seed lots (Meriles, Lamarque, Labuckas, & Maestri, 2004). Matthäus et al. (2004) found increased protein and amino acid contents in winter wheat inoculated with *F. culmorum* compared to an uninoculated control. The reasons for the contradictory results are unknown. Hermann, Aufhammer, Kübler, and Kaul (1999) found no effect of *Fusarium* infection on protein content or other quality parameters in winter wheat, rye, or triticale. However, disease severity was rather low in the latter study and thus some differences between infected and sound grain could have been too small to be detected. Rudgard and Wheeler (1985) reported that the amounts of proline and alanine were positively associated with infection of Brussels sprouts by *Erysiphe cruciferarum* and those of glutamic acid and leucine were negatively associated with infection, indicating that fungal infections can cause changes in the amino acid composition of plants. Hamzehzarghani et al. (2005) observed decreased levels of proline and glycine as a result of *F. graminearum* inoculation in susceptible wheat cultivar Roblin and resistant cultivar Sumai 3. Plant pathogens produce enzymes that cleave proteins to amino acids (i) to inactivate defence response compounds of plants (e.g. cutinases; Murphy, Cameron, Huang, & Vinopal, 1999), (ii) to obtain amino acids for nutrition (Dobinson, Lecomte, & Lazarovits, 1997), or (iii) to degrade host cell walls to facilitate penetration into the host tissue (Dow, Davies, & Daniels, 1998). It was the purpose of this study to test how far *Fusarium* infections can alter the contents of amino acids in wheat grain and to derive a hypothesis about which metabolic processes in the host plants could have been affected by *Fusarium* infection.

2. Materials and methods

2.1. Plant material and fungal isolates

Winter wheat (*Triticum aestivum* L., susceptible cultivar Ritmo) was grown in northern Germany in 2006 near Kiel (latitude 54° 21′ N, longitude 10° 28′ E) and in 2007 near Heide (latitude 54° 12′ N,

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longitude 9° 6′ E). The field experiments were originally carried out to assess different fungicide regimes. For the present study, only grain obtained from control plots (no fungicide use) was utilised. The experimental design was a completely randomised block design with three replicates at Kiel and two replicates at Heide. Plants were inoculated at anthesis during light rainfall by overhead application of a *F. graminearum* macroconidial suspension containing spores of six isolates, as described by Beyer, Klix, and Verreet (2007). Briefly, fungal material was propagated in sterile containers in PDB medium and applied using a plot sprayer (Baumann Saatzucht, Waldenburg, Germany) equipped with standard nozzles (Agrotop, Obertraubling, Germany) such that whole plots were inoculated. Plot size was 10 m^2 (2 × 5 m). Grain was harvested using a combine harvester for experimental plots. Approximately 8 kg of grain were obtained per plot.

Small and light kernels (including high levels of FDKs) were separated from heavy and large kernels (largely excluding FDKs) using a sample cleaner (model SLN, Zuther GmbH, Karwitz, Germany). Furthermore, sand, weed seed and other contaminants were removed during the cleaning process. Four levels of *Fusarium*-damage were generated by mixing varying amounts of light and heavy kernels. Each subsample was mixed well and approximately 200 g of grain were transferred into the laboratory for following analyses. The percentage of FDK was determined based on kernel colour and the degree of shrivelling (Jackowiak, Packa, Wiwart, & Perkowski, 2005; Sinha & Savard, 1997) for each lot of grain, using subsamples of at least 40 kernels. Disease severity levels were 0 ± 2%, 17 ± 2%, 19 ± 2%, and 28 ± 2% FDK for sampling site Kiel and 0 ± 1%, 9 ± 1%, 13 ± 0%, and 25 ± 4% FDK for sampling site Heide. Subsamples were mixed well and approximately 20 g were milled for mycotoxin analyses and 180 g were milled for amino acid analyses.

2.2. Deoxynivalenol (DON) analyses

DON contents of freeze-dried (model Gamma 1-20, Christ Gefriertrocknungsanlagen GmbH, Osterode, Germany) grain were determined as described by Beyer et al. (2007). Briefly, dry samples were milled (laboratory mill model MF 10 basic, IKA-Werke, Stauffen, Germany) and sample material (10 g) was extracted using 50 ml MeOH + H_2O (84 + 16 [v/v]) in Erlenmeyer flasks and shaken for 1.5 h at ≈20 °C. Extracts (10 ml) were filtered and 8 ml of filtered extract were cleaned up using MycoSep 227 Trich+ columns (Coring System Diagnostix GmbH, Gernsheim/Rhein, Germany). Cleaned extract (4 ml) was transferred to round-bottom flasks and evaporated to dryness (Rotavapor model R-210, Büchi Labortechnik AG, Flawil, Switzerland). Residues were re-dissolved in 500 μl of aqueous 12.5% MeOH, transferred to glass vials and stored at −70 °C prior to use. DON contents of the extracts were determined by comparison with a certified type B trichothecene mix standard (biopure Referenzsubstanzen GmbH, Tulln, Austria) using an HPLC system consisting of two L-2130 pumps, a diode array detector (model L-2450) and an EZChrom Elite data analysis unit (all devices from VWR International, Darmstadt, Germany). Five μl of extract were injected into a Chromolith RP-18e column adjusted to 20 °C. Elutions were carried out with MeOH and H_2O. Flow rates were 1.9 ml min^{-1} (0–11 min, 18 min) or 3.7 ml min^{-1} (12–17 min). The gradient programme was as follows. 0–10 min: 12.5% MeOH, 10–12 min: linear gradient from 12.5% to 19% MeOH, 12–17 min: 19% MeOH, 17–18 min: linear gradient from 19% to 12.5% MeOH. Chromatograms were analyzed at 220 nm.

2.3. Determination of amino acids

Contents of amino acids within dry grain samples were determined, following the official protocol given in directive 98/64/EC of the European Commission (Anonymous 1998) in detail.

2.4. Statistics

The number of replicates per disease severity level was $n = 2$ at sampling site Heide and $n = 3$ at sampling site Kiel. Data are presented as median ± standard error. Since the variability of amino acid contents was very small (average coefficient of variation = 1.57%), standard errors of amino acid data were omitted for clarity in Table 1. Regressions were carried out using all data points, not treatment medians or means, and the statistical software package SPSS (version 14, SPSS, Inc., Chicago, Illinois, USA).

3. Results

The percentages of FDK ranged from 0% to 33% at sampling site Kiel and from 0% to 28% at sampling site Heide. DON contents at Kiel were higher than at Heide, even at the same level of disease severity (Fig. 1). For instance, at 20% FDK, DON contents were estimated to be 7.28 mg kg^{-1} at Kiel and 5.01 mg kg^{-1} at Heide (Fig. 1).

The contents of proteinogenic amino acids changed with the percentage of FDK, as well as with the DON content of wheat grain (Table 1). However, significance of the effects depended on the environment. At Kiel, alanine, lysine, and tyrosine contents significantly increased with the DON content or the percentage of FDK whereas arginine, cysteine, glutamic acid, glycine, histidine, isoleucine, leucine, methionine, phenylalanine, proline, serine and valine contents decreased (Table 2). Aspartic acid and threonine were not related to the percentage of FDK or DON contents (Table 2). At Heide, alanine, arginine, lysine, and tyrosine contents significantly increased with the DON content or the percentage of FDK, whereas the glutamic acid level decreased (Table 2). Other amino acids were not significantly affected by *Fusarium* infection at Heide.

Table 1
Sampling sites, percentages of *Fusarium*-damaged kernels (FDK), deoxynivalenol (DON) content, and contents of selected amino acids of winter wheat cultivar Ritmo

Sampling site	FDK (%)	DON (mg kg^{-1})	Amino acid content (% [w/w])[a]															
			ALA	ARG	ASP	CYS	GLU	GLY	HIS	ILE	LEU	LYS	MET	PHE	PRO	SER	THR	TYR	VAL
Kiel	0 ± 2	1.49 ± 0.42	0.45	0.61	0.70	0.29	3.69	0.51	0.33	0.43	0.85	0.35	0.20	0.59	1.24	0.59	0.37	0.33	0.53
Kiel	17 ± 2	3.54 ± 0.62	0.46	0.59	0.70	0.28	3.52	0.50	0.28	0.41	0.82	0.35	0.19	0.57	1.17	0.57	0.37	0.31	0.52
Kiel	19 ± 2	7.49 ± 1.13	0.46	0.58	0.71	0.28	3.42	0.49	0.28	0.41	0.82	0.36	0.19	0.56	1.13	0.56	0.37	0.33	0.52
Kiel	28 ± 2	20.2 ± 4.33	0.46	0.57	0.71	0.27	3.22	0.48	0.27	0.40	0.79	0.36	0.19	0.56	1.09	0.54	0.37	0.35	0.50
Heide	0 ± 1	0.52 ± 0.02	0.44	0.60	0.73	0.29	3.65	0.50	0.29	0.42	0.84	0.36	0.20	0.57	1.18	0.58	0.37	0.31	0.42
Heide	9 ± 1	1.87 ± 1.31	0.44	0.59	0.71	0.29	3.63	0.49	0.28	0.41	0.83	0.35	0.19	0.57	1.20	0.57	0.36	0.32	0.52
Heide	13 ± 0	1.90 ± 0.39	0.44	0.60	0.72	0.29	3.68	0.50	0.29	0.42	0.83	0.36	0.19	0.57	1.17	0.58	0.37	0.31	0.53
Heide	25 ± 4	8.36 ± 3.15	0.45	0.61	0.72	0.29	3.59	0.50	0.29	0.42	0.83	0.37	0.20	0.57	1.17	0.58	0.37	0.34	0.54

The number of replicates was $n = 3$ at sampling site Kiel and $n = 2$ at sampling site Heide. Data represent medians.
[a] ALA = alanine, ARG = arginine, ASP = aspartic acid, CYS = cysteine, GLU = glutamic acid, GLY = glycine, HIS = histidine, ILE = isoleucine, LEU = leucine, LYS = lysine, MET = methionine, PHE = phenylalanine, PRO = proline, SER = serine, THR = threonine, TYR = tyrosine, VAL = valine.

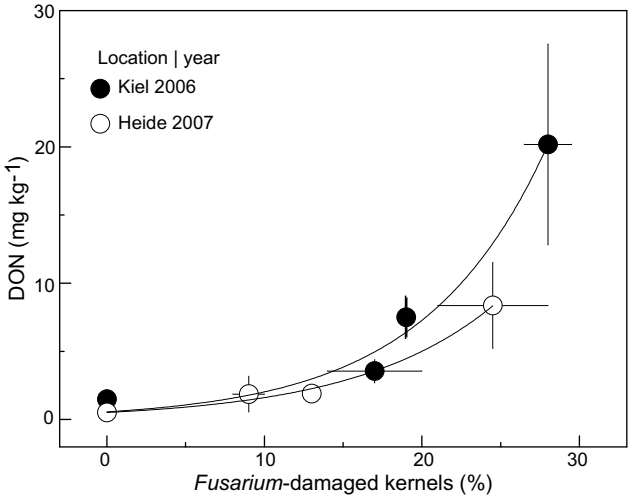

Fig. 1. Relationship between the percentage of *Fusarium*-damaged kernels (FDK) and the deoxynivalenol (DON) content of winter wheat (cv. Ritmo) grain sampled at Kiel in 2006 or at Heide in 2007. Data represent medians ± standard errors.

Amino acid contents of sound kernels were estimated by extrapolating the relationships between amino acid contents and the percentage of FDK to 0. On average, amino acid contents decreased by 0.13% compared to the amino acid content of sound kernels upon an increase of one percentage of FDK.

4. Discussion

4.1. General

DON contents of grain were different among environments, even at the same level of disease severity, expressed as percentage of FDK. *Fusarium*-damaged kernels from Kiel seemed to be more severely affected by the fungus than were those obtained from Heide. Effects of *Fusarium*-damage on amino acid contents of the grain were rather small and, with many amino acids, only significant in the environment with the higher degree of damage. It looks as though *Fusarium*-damage needs to be rather severe to allow the detection of the small changes in amino acid contents caused by the fungal colonization of the kernels.

4.2. Considerations of host–pathogen interaction

Plant pathogens may affect the amino acid composition of their host plant by various mechanisms (van Adel, 1966). First, they may use different amino acids to different extents for their nutrition, thereby decreasing levels of amino acids to different extents. This hypothesis does not explain why contents of some amino acids increased, which would not be the case if consumption of host amino acids by the fungus was the only factor. Second, fungal infection may induce the production of increased levels of those amino acids that can make a contribution to the defence of the plant against the fungus, for instance amino acids needed for the synthesis of proline- or glycine-rich pathogenesis-related proteins (Li et al., 1999). Furthermore, proteins may be decomposed to amino acids by fungal proteolytic enzymes, resulting in lower levels of proteins and higher levels of amino acids in FDKs than in sound kernels. This hypothesis does not explain the decreased levels of some amino acids observed in the present study. Third, fungal infections may alter or even disturb anabolic and/or catabolic processes of the host metabolism resulting in increasing levels for some amino acids, no effect, or decreasing levels for others. For instance, trichothecenes inhibit eukaryotic protein biosynthesis. Hence, increased levels of amino acids, the building units of proteins, may be expected in FDKs compared to sound grain. Furthermore, a combination of the hypotheses could be responsible for the effects observed in the present study. The small size of the effects observed in the present study may be because factors increasing and decreasing amino acid contents acted at the same time. For instance, if proteins are cleaved by fungal enzymes and the resulting amino acids are subsequently metabolized by the fungus, no changes in amino acid level will be detected. However, based on the activity of trichothecenes and proteolytic enzymes, an increase in amino acid contents can be expected, due to *Fusarium*-damage, whereas a decrease can be expected based on the nutritional requirements of the fungus. We tried to elucidate what the amino acids that increased with *Fusarium*-damage had in common and what separated them from the amino acids that decreased or remained constant. The results of those deliberations are summarized below.

Table 2
Changes in amino acid contents (% [w/w]) associated with an increase of one mg DON per kg grain or one percent of *Fusarium*-damaged kernels (FDK) at sampling site Kiel in 2006 or sampling site Heide in 2007

Amino acid	Kiel 2006				Heide 2007			
	DON	R^2	FDK	R^2	DON	R^2	FDK	R^2
Alanine	6.14×10^{-4}	0.47*	3.19×10^{-4}	0.21	1.12×10^{-3}	0.61*	3.10×10^{-4}	0.31
Arginine	-1.07×10^{-3}	0.43*	-1.11×10^{-3}	0.75**	2.40×10^{-3}	0.59*	6.23×10^{-4}	0.26
Aspartic acid	7.49×10^{-4}	0.32	4.35×10^{-4}	0.18	8.01×10^{-4}	0.07	-1.61×10^{-4}	0.02
Cysteine	-9.33×10^{-4}	0.78**	-6.88×10^{-4}	0.69**	8.84×10^{-5}	0.01	-2.13×10^{-5}	0.00
Glutamic acid	-1.97×10^{-2}	0.80**	-1.46×10^{-2}	0.71**	-8.73×10^{-3}	0.50*	-2.21×10^{-3}	0.21
Glycine	-9.91×10^{-4}	0.58**	-9.15×10^{-4}	0.80**	3.20×10^{-4}	0.03	-7.74×10^{-6}	0.00
Histidine	-2.32×10^{-3}	0.46*	-2.36×10^{-3}	0.78**	3.26×10^{-4}	0.05	1.16×10^{-4}	0.04
Isoleucine	-1.25×10^{-3}	0.73**	-1.04×10^{-3}	0.82**	2.63×10^{-4}	0.05	8.13×10^{-5}	0.03
Leucine	-2.48×10^{-3}	0.76**	-2.11×10^{-3}	0.90**	3.74×10^{-4}	0.05	-7.16×10^{-5}	0.01
Lysine	6.52×10^{-4}	0.52**	3.13×10^{-4}	0.20	2.23×10^{-3}	0.71*	7.68×10^{-4}	0.55*
Methionine	-5.44×10^{-4}	0.60**	-4.45×10^{-4}	0.65**	5.78×10^{-4}	0.22	7.35×10^{-5}	0.02
Phenylalanine	-1.25×10^{-3}	0.50*	-1.26×10^{-3}	0.83**	2.72×10^{-4}	0.08	1.80×10^{-4}	0.24
Proline	-6.64×10^{-3}	0.78**	-5.11×10^{-3}	0.75**	-9.74×10^{-4}	0.06	-3.99×10^{-4}	0.07
Serine	-2.18×10^{-3}	0.74**	-1.86×10^{-3}	0.89**	2.23×10^{-4}	0.02	-8.52×10^{-5}	0.02
Threonine	1.51×10^{-5}	0.00	-8.06×10^{-5}	0.03	5.35×10^{-4}	0.15	2.61×10^{-4}	0.24
Tyrosine	1.22×10^{-3}	0.49*	4.02×10^{-4}	0.09	2.33×10^{-3}	0.45	1.07×10^{-3}	0.62*
Valine	-1.09×10^{-3}	0.69**	-8.45×10^{-4}	0.67**	7.53×10^{-3}	0.12	4.45×10^{-3}	0.28

The slope of linear regressions (dependent variable: amino acid, independent variable: DON content or percentage of FDK) was used as estimate for the changes in amino acid contents as affected by DON or FDK. R^2 = coefficient of determination. Significance at the 5% or 1% level is indicated by *, or **, respectively.

4.3. A catabolic-oriented hypothesis

Proteinogenic amino acids are, in a wider sense, products of glycolysis. During glycolysis, glucose-6-phosphate is metabolized to fructose-6-phosphate and subsequently to glycerinaldehyde-3-phosphate on the one hand and dihydroxyacetonephosphate on the other (Fig. 2). Later on, phosphoenolpyruvate and pyruvate are produced. Glyceraldehyde-3-phosphate is the precursor of cysteine, glycine, and serine. All of these amino acids decreased at sampling site Kiel due to *Fusarium* infections (Tables 1 and 2). Furthermore, histidine – a product of the glucose-6-phosphate-derived 6-phosphogluconate – contents decreased due to *Fusarium* infections at Kiel but not at Heide. However, contents of other products of glycolysis that are formed later on remained essentially constant (for instance aspartic acid and threonine), suggesting that the glycolytic input of the metabolic pathway, leading to the production of histidine, remained largely unaffected. Phosphoenolpyruvate and erythrose-4-phosphate are the precursors of phenylalanine and tyrosine. Tyrosine contents increased with the percentage of FDK, while phenylalanine contents decreased at Kiel, but not at Heide. The increase in tyrosine contents suggests that the process of glycolysis was not inhibited, or even stimulated, up to this point. Furthermore, the contents of alanine and lysine, both of them products of pyruvate, increased, irrespective of sampling site, supporting the view that glycolysis was not inhibited or even stimulated. Pyruvate is also further processed in the citric acid cycle. In the citric acid cycle, α-ketoglutarate and oxaloacetate are produced. Oxaloacetate is the precursor of aspartic acid (Fig. 2), an amino acid that was not affected by *Fusarium* infection (Tables 1 and 2). This suggests that the citric acid cycle was not significantly affected. In contrast, contents of glutamic acid, that is derived from α-ketoglutarate, consistently decreased with the DON content or the percentage of FDK, suggesting that *Fusarium* infections either impaired the conversion of α-ketoglutarate to glutamic acid or that the fungus preferentially consumed this amino acid. Effects on amino acids derived from glutamic acid were less clear. Proline

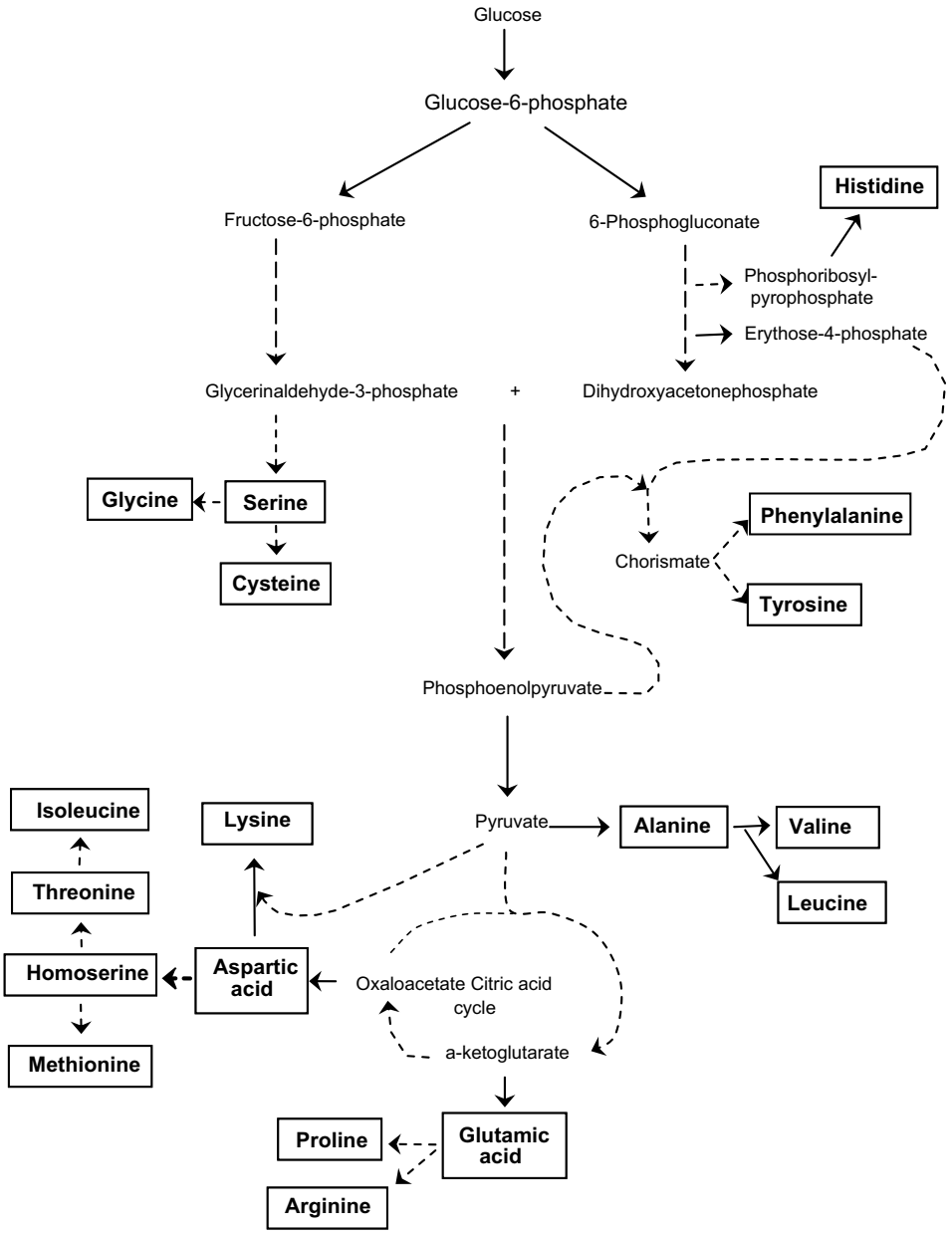

Fig. 2. Biosynthetic pathways of selected proteinogenic amino acid according to Kleber and Schlee (1987). (—) Direct pathway. (- - -) Intermediate steps omitted for clarity.

contents decreased at Kiel, but not at Heide and arginine contents decreased at Kiel but increased at Heide. Contents of leucine and valine, amino acids that are derived from alanine, remained essentially constant or decreased with the degree of *Fusarium*-damage. Levels of amino acids derived from aspartic acid remained essentially constant or decreased slightly.

Effects of *Fusarium* infection on amino acid contents of winter wheat kernels were rather small. With many amino acids, effects were only statistically significant in the environment with the higher level of damage. Besides the small size of the effects, this suggests that infections with *F. graminearum* impaired the production of amino acids derived from glycerinaldehyde-3-phosphate in wheat. Instead, those resources may have been used to produce more phosphoenolpyruvate and pyruvate, resulting in the increased contents of alanine, lysine, and tyrosine that were observed consistently. Furthermore, *Fusarium* infections were associated with decreased levels of glutamic acid.

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References

Anonymous (1998). Commission directive 98/64/EC establishing Community methods of analysis for the determination of amino acids, crude oils and fats, and olaquindox in feedingstuffs and amending directive 71/393/EEC. *Official Journal of the European Communities, L257*, 14–28.

Beyer, M., Klix, M. B., Klink, H., & Verreet, J.-A. (2006). Quantifying the effects of previous crop, tillage, cultivar and triazole fungicides on the deoxynivalenol content of wheat grain - a review. *Journal of Plant Diseases and Protection, 113*(6), 241–246.

Beyer, M., Klix, M. B., & Verreet, J.-A. (2007). Estimating mycotoxin contents of *Fusarium*-damaged winter wheat kernels. *International Journal of Food Microbiology, 119*(3), 153–158.

Beyer, M., Röding, S., Ludewig, A., & Verreet, J.-A. (2004). Germination and survival of *Fusarium graminearum* macroconidia as affected by environmental factors. *Journal of Phytopathology, 152*(2), 92–97.

Beyer, M., Verreet, M. B. J.-A., & Ragab, J.-A. W. S. M. (2005).Effect of relative humidity on germination of ascospores and macroconidia of Gibberella zeae and deoxynivalenol production. *International Journal of Food Microbiology, 98*(3), 233–240.

Bondy, G. S., & Pestka, J. J. (2000). Immunomodulation by fungal toxins. *Journal of Toxicology and Environmental Health, Part B, 3*(2), 109–143.

Dobinson, K. F., Lecomte, N., & Lazarovits, G. (1997). Production of an extracellular trypsin-like protease by the fungal plant pathogen *Verticillium dahliae*. *Canadian Journal of Microbiology, 43*(3), 227–233.

Dow, J. M., Davies, H. A., & Daniels, M. J. (1998). A metalloprotease from *Xanthomonas campestris* that specifically degrades proline/hydroxyproline-rich glycoproteins of the plant extracellular matrix. *Molecular Plant-Microbe Interactions, 11*(11), 1085–1093.

Hamzehzarghani, H., Kushalappa, A. C., Dion, Y., Rioux, S., Comeau, A., Yaylayan, V., et al. (2005). Metabolic profiling and factor analysis to discriminate quantitative resistance in wheat cultivars against fusarium head blight. *Physiological and Molecular Plant Pathology, 66*(4), 119–133.

Hermann, H. W., Aufhammer, A. C. W., Kübler, Y. E., & Kaul, S. H.-P. (ml_chg_old>Hamzehzarghani Hermann et al., 1999). Effects of ear infection with *Fusarium graminearum* on grain quality of winter wheat, winter triticale and winter rye. *Pflanzenbauwissenschaften, 3*(2), 82–87.

Jackowiak, H., Packa, D., Wiwart, M., & Perkowski, J. (2005). Scanning electron microscopy of *Fusarium* damaged kernels of spring wheat. *International Journal of Food Microbiology, 98*(2), 113–123.

Kleber, H.-P., & Schlee, D. (1987). *Biochemie Teil I*. Jena: Gustav Fischer Verlag.

Li, W. L., Faris, J. D., Chittoor, J. M., Leach, J. E., Hulbert, S. H., Liu, D. J., et al. (1999). Genomic mapping of defense response genes in wheat. *Theoretical and Applied Genetics, 98*(2), 226–233.

Matthäus, K., Dänicke, S., Vahjen, W., Simon, O., Wang, J., Valenta, H., et al. (2004). Progression of mycotoxin and nutrient concentrations in wheat after inoculation with *Fusarium culmorum*. *Archives of Animal Nutrition, 58*(1), 19–35.

Meriles, J. M., Lamarque, A. L., Labuckas, D., & Maestri, D. M. (2004). Effect of fungal damage by *Fusarium* spp. and *Diaporthe*/*Phomopsis* complex on protein quantity and quality of soybean seed. *Journal of the Science of Food and Agriculture, 84*(12), 1594–1598.

Murphy, C. A., Cameron, J. A., Huang, S. J., & Vinopal, R. T. (1999). Inactivation of polycaprolactone depolymerase (cutinase) in *Fusarium* cultures by an extracellular protease. *Journal of Industrial Microbiology and Biotechnology, 22*(2), 71–77.

Nightingale, M. J., Marchylo, B. A., Clear, R. M., Dexter, J. E., &K.R. Preston (1999). *Fusarium* head blight: Effect of fungal proteases on wheat storage proteins. *Cereal Chemistry, 76*(1), 150–158.

Rotter, B. A., Prelusky, D. B., & Pestka, J. J. (1996). Toxicology of deoxynivalenol (vomitoxin). *Journal of Toxicology and Environmental Health, 48*(1), 1–34.

Rudgard, S. A., & Wheeler, B. E. J. (1985). The development of *Erysiphe cruciferarum* on field-grown brussels sprouts and associated changes in soluble amino-acids in foliage leaves. *Plant Pathology, 34*(4), 616–625.

Sinha, R. C., & Savard, M. E. (1997). Concentration of deoxynivalenol in single kernels and various tissues of wheat heads. *Canadian Journal of Plant Pathology, 19*(1), 8–12.

Speijers, G. J. A., & Speijers, M. H. M. (2004). Combined toxic effects of mycotoxins. *Toxicology Letters, 153*(1), 91–98.

van Adel, O. M (1966). Amino acids and plant diseases. *Annual Review of Phytopathology, 4*(4), 349–368.