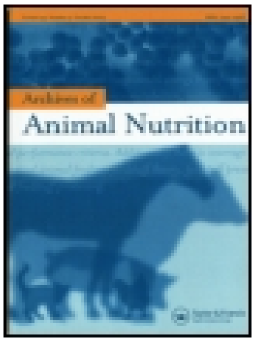

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Variation in chemical composition and physical characteristics of cereal grains from different genotypes

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ABSTRACT

Genotypes of cereal grains, including winter barley (n = 21), maize (n = 27), oats (n = 14), winter rye (n = 22), winter triticale (n = 21) and winter wheat (n = 29), were assayed for their chemical composition and physical characteristics as part of the collaborative research project referred to as GrainUp. Genotypes of one grain species were grown on the same site, except maize. In general, concentrations of proximate nutrients were not largely different from feed tables. The coefficient of variation (CV) for the ether extract concentration of maize was high because the data pool comprised speciality maize bred for its high oil content. A subset of 8 barley, 20 rye, 20 triticale and 20 wheat samples was analysed to differ significantly in several carbohydrate fractions. Gross energy concentration of cereal grains could be predicted from proximate nutrient concentration with good accuracy. The mean lysine concentration of protein was the highest in oats (4.2 g/16 g N) and the lowest in wheat (2.7 g/16 g N). Significant differences were also detected in the concentrations of macro elements as well as iron, manganese, zinc and copper. Concentrations of arsenic, cadmium and lead were below the limit of detection. The concentration of lower inositol phosphates was low, but some inositol pentaphosphates were detected in all grains. In barley, relatively high inositol tetraphosphate concentration also was found. Intrinsic phytase activity was the highest in rye, followed by triticale, wheat, barley and maize, and it was not detectable in oats. Substantial differences were seen in the thousand seed weight, test weight, falling number and extract viscoelasticity characteristics. The study is a comprehensive overview of the composition of different cereal grain genotypes when grown on the same location. The relevance of the variation in composition for digestibility in different animal species will be subject of other communications.

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1. Introduction

Many studies have been conducted to determine the chemical composition and physical characteristics of cereal grains used in livestock feeding. Results of these studies are part of comprehensive feed tables (e.g. DLG 1997, c2006–2010; NRC 2012; Agroscope c2011–2015). These feed tables indicate variations in chemical composition, such as concentrations of proximate nutrients, starch, amino acids, minerals and energy between and within cereal grain species. Information on the physical characteristics of cereal grains, such as thousand seed weight (TSW), test weight (TW) and falling number (FN), or on the content of inositol phosphates, intrinsic phytase activity and specific carbohydrate fractions is scarce. Little is known about how environmental and genetic factors contribute to variations in the chemical composition and physical characteristics of cereal grains. Year of harvest, rainfall, temperature, soil conditions, fertilisation and other agronomic details, as well as harvesting and storage conditions, can affect chemical characteristics of the cereal grains, including their energy content, starch, crude protein, fibre fractions or minerals (Longstaff and McNab 1986; Conan et al. 1992; Zebarth et al. 1992; Metayer et al. 1993). Moreover, due to progress in breeding of cereal grains, alterations in content of proximate nutrients, particularly in crude protein (CP) concentration and amino acid composition, have recently been reported (e.g. Murphy et al. 2009; Peltonen-Sainio et al. 2012). Thus, characterisation of the variations in nutritional value of cereal grains that result from genotypic differences may help in defining appropriate breeding objectives for improving the feeding value of cereal grains for livestock nutrition.

The main objective of this work was to study the chemical constituents of different genotypes of cereal grains and their variation. Genotypes of winter barley, maize, oats, winter rye, winter triticale and winter wheat were assayed for their chemical composition and physical characteristics. Analyses included the content of the proximate nutrients, fibre fractions, carbohydrate fractions, Klason lignin, amino acids, energy, minerals, inositol phosphates and intrinsic phytase activity, as well as TSW, TW and FN. Apart from maize, plants with different genotypes were grown and harvested under the same standardised agronomic conditions in field plots in the quantities required to conduct digestibility studies in different animal species. The study was a central element of the collaborative research project referred to as GrainUp (www.grain-up.de).

2. Materials and methods

2.1. *Cultivation and processing of cereal grains*

Several different cereal grains were investigated including different genotypes of winter barley (n = 21), maize (n = 27), oats (n = 14), winter rye (n = 22), winter triticale (n = 21) and winter wheat (n = 29). Genotypes were chosen to represent previously known differences in CP concentration and yield. The grains were grown at different locations of the Agriculture Experiment Station of the University of Hohenheim, Stuttgart, Germany. To the extent possible, genotypes of each species were grown at a single plot with the exception of maize. To obtain genuine allogamous maize seed, production had to be carried out in isolation. Because of limited availability of isolated plots, the seeds of only four varieties could be produced at the Eckartsweier location of the Experiment station itself. The seeds of another four varieties were harvested from

Table 1. Description of the experimental locations.

	Meiereihof	Heidfeldhof	Eckartsweier
Species grown	Barley, rye, triticale, wheat	Oats	Maize
Altitude above sea level [m]	350	400	141
Average annual rainfall [mm]	700	697	726
Average temperature [°C]	8.8	8.8	9.9
Soil type	Sandy clay loam	Silty loam with stones	Loamy sand to loamy clay
Ground points	65–74	55–60	35–80
pH value	6.8	6.7	6.7
P_2O_5 [mg/100 g soil]	33	15	21
K_2O [mg/100 g soil]	34	19	13
MgO [mg/100 g soil]	13	16	9
Humus [mg/100 g soil]	2.3	1.8	–

the centre of fields owned by different farmers in the Eckartsweier area. For the remaining 19 genotypes, genuine seed was obtained in adequate quantities from the respective breeding companies. A description of the field sites including the soil and agronomic conditions is given in Table 1.

Grain samples of each genotype were obtained at their respective times of ripening. Because of rainy conditions during the period of harvest, grains were gently dried to lower the moisture content to levels of about 12%. After drying, the cereal grains were sieved to remove straw residues and very small seeds, and then stored at about 4°C.

2.2. *Chemical analyses of cereal grains*

For chemical analyses, all grain samples were ground through a sieve with a pore size of 0.5 mm (Siebtechnik GmbH, Mühlheim-Ruhr, Germany, and Retsch GmbH, Haan, Germany) if not otherwise specified. Hulled genotypes of barley and oats were not dehulled before grinding. With the exception of maize, a vibrating cup mill (Type 6-TOPF, Siebtechnik GmbH, Mülheim-Ruhr, Germany) was used to grind samples for inositol phosphate and phytase analyses. Ground samples were stored in a freezer prior to analysis of inositol phosphates and phytase.

The following chemical analyses were conducted: dry matter (DM) (method 3.1), crude ash (CA) (method 8.1), crude protein (CP) (method 4.1.1), crude fibre (CF) (method 6.1.1), ether extract (EE) (method 5.1.1b), starch (method 7.2.1), neutral detergent fibre (aNDFom) (6.5.1), acid detergent fibre (ADFom) (6.5.2), acid detergent lignin (ADL) (6.5.3) and the minerals Ca, P, Mg, K, Na, Fe, Mn, Zn and Cu (methods 10 and 11) according to official methods (Verband Landwirtschaftlicher Untersuchungs- und Forschungsanstalten (VDLUFA) 2007). Gross energy (GE) was determined using a bomb calorimeter (C 200; Ika-Werke GmbH & Co. KG, Staufen, Germany). Carbohydrates, low-molecular sugars, fructans, soluble and insoluble β-glucan, total non-starch polysaccharides (NSP) divided into soluble and insoluble non-cellulosic polysaccharides and cellulose, and Klason lignin concentrations were determined according to Bach Knudsen (1997) in a subset of 8 barley, 20 rye, 20 triticale and 20 wheat samples. Soluble, insoluble and total arabinoxylans were determined from the sum of arabinose and xylose residues in the soluble, insoluble and total NSP fractions. For amino acid (AA) analysis, samples were oxidised using performic acid and hydrogen peroxide (Rodehutscord et al. 2004) and then hydrolysed with 6 M HCl for 24 h at 110°C. Norleucine was used as the internal standard.

Separation and detection of AA was done on an AA analyser (L8900, VWR/Hitachi). Photometric detection was done at 570 nm (440 nm for proline) with post-column ninhydrin derivatisation. Methionine and cysteine were determined as methionine sulphone and cysteic acid, respectively. Tryptophan was determined by reversed-phase chromatography and fluorescence detection after alkaline hydrolysis, using barium hydroxide according to Scheuermann and Eckstein (1986) using an Agilent 1100 HPLC (Agilent, Waldbronn, Germany). A Nucleosil 120 5 C18 column (125 X 4) with a corresponding guard column was used as stationary phase. The mobile phase consisted of a mixture of 0.01 M sodium acetate buffer (pH 4.5)/methanol (86/14 (v/v)), flow rate was 0.8 ml/min, column temperature 20°C. All AA concentrations were expressed as g/16 g N. Phytase activity in the grains was determined according to Greiner and Egli (2003) (method 2: direct incubation). Activity was expressed in units (U), whereby the unit was defined as 1 µmol of phosphate liberated from 100 µmol potassium phytate per minute at 45°C, pH 5.0. Phytic acid (*myo*-inositol 1,2,3,4,5,6-hexakis (dihydrogen phosphate); $InsP_6$) and other inositol phosphate isomers ($InsP_x$) were measured by high performance ion exchange chromatography (Dionex ICS-3000, Idstein, Germany, using a CarboPac® PA 200 column) with post-column derivatisation, following extraction with 0.2 M ethylenediaminetetraacetic acid (EDTA) and 0.1 M sodium fluoride at pH 10 as described by Zeller et al. (2015).

2.3. *Physical characteristics of cereal grains*

The TSW of each genotype was determined according to the method described by the International Seed Testing Association (ISTA 2013). The TW is given as kilograms per hectolitre of grain, and was calculated from a measured quarter of a litre of grain material. The FN was measured on two subsamples of 7 g of wholemeal for barley, oats, rye and wheat, and a sample of 9 g of wholemeal for triticale, based on the method of the International Association for Cereal Chemistry (IACC 1968).

For extract viscoelasticity measurements, grain samples were ground through a 1-mm sieve size using a FOSS Cyclotec 1093 mill (Foss Tecator AB, Höganäs, Sweden). Ten grams of each grain sample were mixed with 40 ml of distilled water in a 300 ml Erlenmeyer flask (G20, Schott AG, Mainz, Germany) and immediately incubated in a shaking water bath at 39°C for 30 min. Samples were then centrifuged at 39°C for 2 min at 10,000 × *g*. The supernatant was instantly pipetted out and homogenised. Forty minutes after the incubation started, 7.5 ml of the homogenised supernatant were used to fill the measuring unit of a rheometer (Physica MCR300 with a DG26.7 double gap measuring system). The filled measuring unit was preheated to 39°C and then measurements were recorded at 39°C ± 0.1°C. One hundred measurements were recorded logarithmically for a range of shear rates from 0.1 to 1000 s^{-1} at intervals of 2.5 s. Each grain sample was measured in duplicate. Extract viscoelasticity analysis of each sample followed the Herschel–Bulkley rheological model, calculated as $\tau = \tau_0 + k\dot{\gamma}^n$, where τ is the shear stress [mPa], τ_0 is the yield point [mPa], k is the consistency index [mPa · s^n], $\dot{\gamma}$ is the shear rate [s^{-1}] and n is the flow index (dimensionless). For each sample, shear stress values after the last negative value were included. Extract viscoelasticity regression was computed using OriginPro 8.1 (OriginLab Corporation, Northampton, MA, USA).

2.4. *Statistical analyses*

For each cereal grain, minimum (Min), maximum (Max) and mean values as well as standard deviations (SD) and coefficient of variation (CV) were determined using the PROC MEANS procedure of SAS (2008; Inst. Inc., Cary, NC). The symbol "•" is used in the tables to indicate that analysed values are above the limit of detection but below the limit of quantification of the respective method. In such case, the average value between the limit of detection and the limit of quantification was used for statistical analysis. If any analysed values were below the limit of detection, they have been mentioned in the tables as "n.d.". Standard deviations were only included when more than 50% of the analysed values were above the limit of quantification. If more than 50% of the analysed value was below the limit of quantification it has been declared in the tables with "Δ". Significant differences between means of cereal grains were determined by pairwise *t*-tests using a general linear model (GLM, SAS). Significant differences have been indicated by different superscripts ($p \leq 0.05$). For each cereal grain, Pearson correlation coefficients between all analysed traits were calculated with the CORR procedure of SAS, but only a few of these correlations are presented and discussed herein.

Given the high importance of GE values in animal feeding and the limitations that institutions may have in determining GE, we tried to estimate the GE of each grain. For estimation of GE, a stepwise selection with the variables CP, CF, EE and nitrogen-free extract (NFE) using PROC GLMSELECT was made for each grain type separately and for all grain types together with the options select = SL, stop = none, choose = AIC. The Akaike information criterion (AIC) was taken as the measure for the goodness of fit in addition to the root mean square error (RMSE).

3. Results and discussion

Tables contain mean values, SD, CV, as well as Min and Max values for each cereal grain. Values of individual genotypes are available online as Supplementary Tables.

3.1. *Proximate nutrients, carbohydrate fractions, lignin and gross energy*

3.1.1. *Crude protein*

The mean CP concentration ranged from 93.5 g/kg DM in maize to 137 g/kg DM in wheat (Table 2). Results were in general agreement with Agroscope (c2011–2015), except for wheat, which contained less CP in the present work. For all cereal grains, variation between genotypes was relatively high, with CV values ranging from 4.3% in rye to 6.8% in wheat.

3.1.2. *Ether extract*

The mean EE concentration ranged from 18.8 g/kg DM in rye to 56.8 g/kg DM in maize and was significantly ($p \leq 0.05$) lower in wheat, triticale and rye than maize and oats, with barley being intermediate. The CV of EE concentration was especially high in oats (14%) and maize (35%). The large variation in EE concentration of maize can be attributed to genetic variations between the maize hybrids analysed, as the assay

Table 2. Concentration of crude nutrients, fibre fractions, starch and gross energy in cereal grains.

		Dry matter [%]	Crude ash [g/kg DM]	Crude protein [g/kg DM]	Crude fibre [g/kg DM]	Ether extract [g/kg DM]	NFE$^{•}$ [g/kg DM]	aNDFom* [g/kg DM]	ADFom$^{\#}$ [g/kg DM]	ADL† [g/kg DM]	Starch [g/kg DM]	Gross energy [MJ/kg DM]
Barley (n = 21)	Mean	88.2^{c}	24.9^{b}	123^{b}	42.2^{b}	28.8^{b}	787^{d}	187^{b}	55.5^{b}	7.67^{b}	616^{e}	18.7^{c}
	Min	87.7	21.6	108	35.2	24.4	743	152	44.2	4.50$^{\bullet}$	567	18.5
	Max	88.5	40.7	136	54.6	34.1	799	208	71.1	9.29	642	19.1
	SD‡	0.23	3.73	7.4	5.48	2.37	12.4	13.4	7.85	1.213	15.6	0.11
	CV¶ [%]	0.26	14.9	5.99	13.0	8.22	1.59	7.16	14.2	15.8	2.54	0.58
Maize (n = 27)	Mean	90.3^{a}	13.3^{e}	93.5^{d}	18.7^{cd}	56.8^{a}	818^{b}	88.9^{f}	27.4^{d}	4.50$^{\bullet c}$	740^{a}	19.2^{b}
	Min	89.6	10.9	78.1	14.0	41.7	740	71.0	21.1	$^{\Delta}$	660	18.8
	Max	91.8	16.4	112	22.0	123	848	110	32.2	$^{\Delta}$	783	20.7
	SD	0.59	1.51	9.18	2.23	19.73	25.7	11.32	2.86	$^{\Delta}$	28.2	0.50
	CV [%]	0.66	11.3	9.82	12.0	34.8	3.14	12.7	10.5	$^{\Delta}$	3.82	2.44
Oats (n = 14)	Mean	89.3^{b}	28.2^{a}	127^{b}	104^{a}	52.0^{a}	689^{e}	289^{a}	129^{a}	20.5^{a}	495^{f}	19.4^{a}
	Min	89.0	25.2	121	88.7	44.8	621	261	111	7.38	410	19.2
	Max	89.9	31.1	141	134	72.9	711	341	167	34.5	538	20.1
	SD	0.25	1.70	5.5	12.2	7.23	22.7	26.2	15.9	7.19	37.2	0.26
	CV [%]	0.28	6.05	4.31	11.8	13.9	3.30	9.04	12.3	35.1	7.52	1.35
Rye (n = 22)	Mean	88.0^{c}	17.2^{c}	117^{c}	17.9^{d}	18.8^{c}	829^{a}	146^{c}	29.6^{cd}	8.58^{b}	643^{d}	18.4^{d}
	Min	87.5	16.2	108	15.8	17.5	819	126	23.8	6.58	631	18.3
	Max	88.4	19.3	127	20.4	20.9	839	172	33.6	11.1	659	18.5
	SD	0.21	0.86	5.0	1.30	0.95	4.96	12.0	2.50	1.255	6.9	0.07
	CV [%]	0.24	5.01	4.28	7.28	5.06	0.60	8.18	8.47	14.6	1.07	0.40
Triticale (n = 21)	Mean	88.0^{c}	18.0^{c}	124^{b}	21.0^{c}	19.1^{c}	818^{b}	134^{d}	28.9^{d}	7.50^{b}	699^{c}	18.4^{d}
	Min	87.6	16.9	113	15.8	16.0	803	101	24.5	4.50$^{\bullet}$	641	18.2
	Max	88.7	19.2	138	25.5	22.7	829	169	33.0	10.0	727	18.5
	SD	0.25	0.63	6.5	2.40	1.89	6.91	19.9	2.52	1.284	18.6	0.08
	CV [%]	0.29	3.52	5.23	11.4	9.93	0.85	14.8	8.72	17.1	2.66	0.42
Wheat (n = 29)	Mean	87.7^{d}	16.1^{d}	137^{a}	21.3^{c}	22.3^{c}	804^{c}	120^{e}	31.4^{c}	7.83^{b}	713^{b}	18.6^{c}
	Min	87.2	14.6	121	18.6	18.7	777	101	24.6	4.50$^{\bullet}$	686	18.4
	Max	88.1	18.5	162	26.2	26.7	817	137	37.7	11.2	735	18.8
	SD	0.21	0.74	9.3	2.03	2.06	10.2	10.0	3.35	1.674	9.8	0.09
	CV [%]	0.24	4.58	6.79	9.55	9.25	1.26	8.33	10.7	21.4	1.38	0.51

Notes: $^{•}$NFE, nitrogen-free extract; *aNDFom, neutral detergent fibre; $^{\#}$ADFom, acid detergent fibre; †ADL, acid detergent lignin; ‡SD, standard deviation; ¶CV, coefficient of variation; $^{\Delta}$Other statistical values were not determined, more than 50% of analysed values were below the limit of quantification; $^{\bullet}$Mean between limit of detection and limit of quantification. $^{a\text{-}f}$Means within a column not showing common superscript letter are significantly different between grain types; All corresponding individual values for each genotype are available online (Tables S1a–1f).

comprised conventional maize hybrids with normal oil content as well as specialty maize bred for high oil content.

3.1.3. *Fibre fractions*

The mean CF concentration ranged from 17.9 g/kg DM in rye to 104 g/kg DM in oats and was significantly different between most of the grains ($p \leq 0.05$). The mean CF values were in general agreement with values reported by Agroscope (c2011–2015), while mean values reported in the DLG feed table were higher, ranging from 25 g/kg DM in maize and rye to 133 g/kg DM in oats. The CV of CF concentration was similar for the grains and ranged from 7.3% in rye to 13% in barley. Mean ADFom values were all slightly above mean CF values, but the differences between cereal grains as well as CV within each grain species were very similar for both criteria. The mean ADL concentration was the highest in oats while it was below the limit of quantification in maize. The ADL concentration did not differ between the other cereal grains with an average of 8 g/kg DM, but the CV of ADL concentration was relatively high for all grains and the highest for oats (35%). Regarding aNDFom, the highest mean concentration was found in oats (289 g/kg DM) and the lowest in maize (88.9 g/kg DM), with significant differences between all cereal grains ($p \leq 0.05$) and substantial variation within each grain (CV between 7.2% and 15%). Mean values were in agreement with Agroscope (c2011–2015).

3.1.4. *Starch and other carbohydrate fractions*

The starch concentration significantly differed between all cereal grains ($p \leq 0.05$) with mean values ranging from 495 g/kg DM in oats to 740 g/kg DM in maize (Table 2). Means found for barley, maize, rye, and triticale corresponded to Agroscope (c2011–2015), while results for oats and wheat were lower in the present work. Values reported by DLG (c2006–2010) were lower overall. In the present study, the starch concentration varied greatly especially between genotypes of oats (CV 7.5%), while the CV was relatively low in the other grains (1.1%–3.8%).

Other carbohydrate fractions and Klason lignin were determined in 20 genotypes of rye, triticale and wheat, and in eight genotypes of barley (Tables 3 and 4). The mean concentration of total NSP ranged from 98.2 g/kg DM in wheat to 172 g/kg DM in barley. The mean proportion of soluble NSP in total NSP was 29%, 42%, 20% and 19% in barley, rye, triticale and wheat, respectively. The mean concentration of the soluble β-glucans and arabinoxylans ranged from 0.9 g/kg DM in triticale to 24.1 g/kg DM in rye, and from 9.7 g/kg DM in barley to 30.9 g/kg DM in rye, respectively. Compared to the total concentrations of β-glucans and arabinoxylans, the CV of the respective soluble parts was higher. In the majority of cases, the CV of the concentrations of the soluble non-cellulosic polysaccharide fractions arabinose, xylose, mannose, galactose, glucose and uronic acid also was higher compared to the concentrations of total fractions. The concentrations of carbohydrate fractions in barley, rye and wheat were in general agreement with the values of Bach Knudsen (1997, 2014), but concentrations of Klason lignin were lower in the present study. In this context, further studies with animals are warranted to evaluate the impact of the observed variations in carbohydrate fractions and lignin on nutrient digestibility.

Table 3. Concentration of carbohydrate fractions and Klason lignin in cereal grains [g/kg DM].

		Low molecular weight carbohydrates					Non-starch polysaccharides (NSP)							
		Glucose	Fructose	Sucrose	Total sugars	Fructans	Cellulose	Total β-glucans	Soluble β-glucans	Total arabinoxylans	Soluble arabinoxylans	Total NSP	Soluble NSP	Klason lignin
Barley	Mean	1.8^{b}	1.1^{c}	14.7^{c}	17.5^{c}	6.0^{c}	27.5^{a}	46.7^{a}	24.1^{a}	77.4^{b}	9.7^{c}	172^{a}	50.6^{a}	22.1^{a}
(*n* = 8)	Min	1.6	0.8	13.4	16.3	5.0	13.9	39.5	20.3	70.5	4.0	167	37.6	15.2
	Max	1.9	1.3	16.9	19.5	6.9	37.0	53.3	27.0	86.4	20.1	184	66.1	27.4
	SD‡	0.10	0.17	1.13	1.15	0.57	7.08	4.50	2.53	4.78	5.29	5.81	9.44	4.15
	CV¶ [%]	6.02	15.6	7.70	6.59	9.17	25.8	9.63	10.5	6.18	54.7	3.38	18.7	18.8
Rye	Mean	6.1^{a}	1.8^{a}	25.8^{a}	33.6^{a}	29.1^{a}	11.9^{c}	20.1^{b}	6.6^{b}	85.4^{a}	30.9^{a}	139^{b}	41.2^{b}	18.2^{b}
(*n* = 20)	Min	3.7	1.2	20.5	28.6	24.7	5.9	16.9	4.9	74.3	24.2	122	32.7	9.4
	Max	8.8	2.4	32.0	37.8	34.5	18.2	26.4	8.6	96.1	40.5	158	53.5	32.3
	SD	1.70	0.38	3.37	2.43	2.65	2.75	2.86	1.05	5.72	3.95	9.30	5.06	5.93
	CV [%]	28.1	21.0	13.1	7.23	9.11	23.2	14.2	16.0	6.70	12.8	6.69	12.3	32.6
Triticale	Mean	5.6^{a}	1.4^{b}	23.2^{b}	30.1^{b}	5.7^{c}	19.3^{b}	6.6^{c}	0.9^{d}	55.3^{d}	12.6^{b}	103^{c}	20.6^{c}	17.0^{b}
(*n* = 20)	Min	3.0	0.9	17.0	23.0	2.1	11.8	5.5	0#	40.2	8.1	91.6	15.2	11.2
	Max	9.7	2.5	31.6	41.0	8.9	27.2	7.7	1.7	73.8	17.4	115	31.1	21.7
	SD	1.90	0.36	4.26	5.47	1.77	4.73	0.68	0.66	12.0	2.53	7.12	3.75	2.52
	CV [%]	34.0	25.9	18.4	18.1	31.1	24.5	10.2	75.2	21.7	20.0	6.89	18.2	14.8
Wheat	Mean	1.9^{b}	0.9^{c}	14.3^{c}	17.1^{c}	9.8^{b}	14.4^{c}	6.1^{c}	2.0^{c}	63.7^{c}	13.9^{b}	98.2^{c}	19.1^{c}	10.8^{c}
(*n* = 20)	Min	1.3	0.5	9.95	12.7	7.1	11.9	4.6	1.3	58.5	8.3	89.5	10.6	6.7
	Max	2.4	1.3	19.5	21.9	14.0	16.4	7.8	3.4	74.2	22.5	113	29.7	15.7
	SD	0.32	0.23	2.48	2.35	1.67	1.42	0.89	0.52	4.20	3.44	6.10	4.57	3.01
	CV [%]	16.8	26.5	17.3	13.8	17.0	9.87	14.5	26.4	6.60	24.7	6.22	24.0	27.9

Notes: ‡SD, standard deviation; ¶CV, coefficient of variation; #Difference of analysed total and insoluble concentration below 0. a–d Means within a column not showing common superscript letter are significantly different between grain types; All corresponding individual values for each genotype are available online (Tables S2a–2d).

Table 4. Concentration of non-cellulosic polysaccharide fractions in cereal grains [g/kg DM].

		Total arabinose	Soluble arabinose	Total xylose	Soluble xylose	Total mannose	Soluble mannose	Total galactose	Soluble galactose	Total glucose	Soluble glucose	Total uronic acid	Soluble uronic acid
Barley	Mean	28.3^{b}	5.4^{b}	49.1^{a}	4.3^{c}	3.7^{c}	1.1^{b}	4.2^{a}	1.9^{a}	55.4^{a}	37.3^{a}	3.5^{a}	0.6^{ab}
(n = 8)	Min	25.4	3.3	45.1	0$^{\#}$	3.3	0.7	3.5	1.3	47.0	27.6	3.3	0.3
	Max	33.5	10.3	53.1	9.9	4.4	1.4	7.1	4.8	68.5	45.1	3.9	0.8
	SD‡	2.60	2.35	2.73	3.2	0.39	0.26	1.20	1.21	6.75	5.74	0.21	0.16
	CV¶ [%]	9.18	43.5	5.56	74.3	10.5	24.7	28.8	64.2	12.2	15.4	5.94	26.5
Rye	Mean	34.9^{a}	12.6^{a}	50.5^{a}	18.4^{a}	5.4^{a}	2.3^{a}	4.5^{a}	1.5^{b}	28.5^{b}	5.8^{b}	2.6^{c}	0.7^{a}
(n = 20)	Min	31.0	10.4	43.3	13.9	4.6	1.7	3.7	1.1	24.5	1.9	2.2	0.5
	Max	40.6	16.1	56.9	24.3	6.5	3.0	5.2	1.7	33.6	7.6	2.9	0.9
	SD	2.39	1.49	3.58	2.51	0.58	0.40	0.37	0.17	2.72	1.25	0.20	0.15
	CV [%]	6.85	11.8	7.09	13.6	10.6	17.7	8.14	11.7	9.55	21.7	7.76	19.8
Triticale	Mean	22.3^{c}	5.2^{b}	32.9^{c}	7.4^{b}	4.3^{b}	1.3^{b}	4.2^{a}	1.9^{a}	17.2^{c}	3.8^{c}	2.8^{bc}	0.7^{ab}
(n = 20)	Min	14.3	3.1	25.9	5.0	2.7	0.6	3.5	1.5	9.8	0.6	2.2	0.4
	Max	31.0	7.4	42.8	10.1	5.8	2.6	5.1	2.3	24.8	11.2	3.3	1.1
	SD	5.73	1.24	6.32	1.36	0.97	0.53	0.45	0.28	4.76	2.16	0.33	0.16
	CV [%]	25.6	23.8	19.2	18.4	22.9	40.2	10.7	14.8	27.7	57.8	11.7	23.6
Wheat	Mean	24.8^{c}	5.45^{b}	39.0^{b}	8.5^{b}	2.4^{d}	0.6^{c}	3.6^{b}	2.0^{a}	10.7^{d}	1.89^{d}	2.9^{b}	0.6^{b}
(n = 20)	Min	22.1	3.34	35.7	5.0	2.0	0.2	3.2	1.6	8.0	0$^{\#}$	2.5	0.2
	Max	27.8	8.15	46.4	14.4	2.9	0.9	4.1	2.3	12.3	3.91	3.4	0.8
	SD	1.81	1.19	2.55	2.29	0.24	0.14	0.19	0.16	1.25	1.17	0.30	0.18
	CV [%]	7.30	21.8	6.55	27.0	9.90	23.5	5.35	7.96	11.7	61.6	10.3	30.0

Notes: ‡SD, standard deviation; ¶CV, coefficient of variation; #Difference of analysed total and insoluble concentration below 0. $^{a–d}$ Means within a column not showing common superscript letter are significantly different between grain types; All corresponding individual values for each genotype are available online (Tables S3a–3d).

3.1.5. *Gross energy*

The mean GE concentration ranged from 18.4 MJ/kg DM in rye and triticale to 19.4 MJ/kg DM in oats and significantly differed between cereal grains ($p \leq 0.05$) except between barley and wheat and between rye and triticale, respectively. Variation within grains was very low (CV < 0.6%) in barley, rye, triticale and wheat. Variation was higher in oats (CV 1.4%) and maize (CV 2.4%).

The variation in GE content between genotypes of maize, oats, barley and triticale may be partially attributable to variation in EE content, as indicated by a positive correlation between GE and EE content (r = 0.48–0.93; $p \leq 0.05$) in these grains (Table S10). A positive correlation with GE was also detected for CP in barley, oats, rye and wheat.

The variation in GE concentration observed, particularly for maize and oats, may be of significance for the formulation of livestock diets, because cereals are major dietary ingredients and the largest contributor to dietary energy. Further studies using animals are warranted to determine whether the observed variation in GE concentration is also reflected in digestible energy, metabolizable energy and net energy values of the respective cereals.

As judged by values for AIC and RMSE, the GE content could be accurately estimated using organic fractions (Table 5). For all grains, CP and NFE were selected as model variables. The stepwise selection also included other fractions in different combinations for each grain type. In the equation for maize, apart from CP and NFE, EE was chosen. These equations may be used to predict GE in cereal grains in cases where analytical determination is not possible.

3.2. *Amino acids*

The mean Lys concentration ranged from 2.7 g/16 g N in wheat protein to 4.2 g/16 g N in oat protein and differed significantly between all cereal grains ($p \leq 0.05$) (Table 6). The CV for the concentration of Lys in protein was especially high in maize (7.7%). Similar differences and variation were seen in the Met concentration of the grain proteins. The mean Met concentration ranged from 1.5 g/16 g N (CV 3.7%) in wheat protein to 2.1 g/16 g N (CV 11%) in maize protein. Significant and considerable differences between grain proteins were also detected in the Thr and Trp concentrations. The mean Thr concentration ranged from 2.9 g/16 g N (CV 2.3%) in wheat protein to 3.7 g/16 g N (CV 1.8%) in maize protein, and the mean Trp concentration from 0.8 g/16 g N (CV 8.5%) in maize protein to 1.4 g/16 g N (CV 4.1%) in oat protein. In general, concentrations of Lys, Met, Thr and Trp were in good agreement with data from DLG (c2006–2010) and Agroscope (c2011–2015). However, the Lys concentration

Table 5. Selected variables and estimated parameters of gross energy (GE) estimation.

	AIC[†]	RMSE[‡]	Equation*
Barley	−94.5	0.056	GE = 0.0245 · CP + 0.0201 · CF + 0.0371 · EE + 0.0176 · NFE
Maize	−49.9	0.089	GE = 0.0248 · CP + 0.0210 · CF + 0.0443 · EE + 0.0171 · NFE
Oats	−48.4	0.220	GE = 0.0234 · CP + 0.0396 · EE + 0.0181 · NFE
Rye	−103	0.054	GE = 0.0262 · CP + 0.0185 · NFE
Triticale	−88.6	0.065	GE = 0.0241 · CP + 0.0137 · CF + 0.0426 · EE + 0.0175 · NFE
Wheat	−104	0.092	GE = 0.0240 · CP + 0.0192 · CF + 0.0185 · NFE
All grains	−415	0.126	GE = 0.0215 · CP + 0.0236 · CF + 0.0395 · EE + 0.0177 · NFE

Notes: [†]AIC, Akaike information criterion; [‡]RMSE, root-mean-square error; *GE in MJ/kg and nutrients in g/kg.

Table 6. Sum of all detected amino acids (AA) and concentration of essential AA in crude protein of cereal grains.

y		Sum of all AA	Arg	His	Ile	Leu	Lys	Met	Phe	Thr	Trp	Val
		[g/kg DM]	[g/16 g N]									
Barley	Mean	118^{bc}	4.87^{c}	2.36^{d}	3.13^{c}	6.75^{c}	3.49^{c}	1.57^{c}	5.12^{a}	3.39^{c}	1.23^{b}	4.42^{b}
(*n* = 21)	Min	104	4.62	2.24	2.88	6.33	3.17	1.49	4.69	3.21	1.13	4.14
	Max	133	5.16	2.59	3.42	7.15	3.85	1.69	5.42	3.59	1.31	4.80
	SD‡	7.5	0.163	0.097	0.147	0.212	0.173	0.059	0.196	0.122	0.053	0.184
	CV¶ [%]	6.33	3.34	4.13	4.69	3.14	4.97	3.77	3.82	3.61	4.28	4.15
Maize	Mean	94.0^{e}	4.63^{d}	3.07^{a}	3.28^{b}	12.6^{a}	2.98^{e}	2.06^{a}	4.95^{b}	3.65^{a}	0.75^{f}	4.49^{b}
(*n* = 27)	Min	77.0	4.12	2.98	2.98	11.1	2.51	1.72	4.51	3.52	0.64	4.14
	Max	116	5.34	3.23	3.49	14.1	3.53	2.69	5.30	3.78	0.89	4.78
	SD	10.23	0.261	0.072	0.129	0.70	0.230	0.225	0.188	0.065	0.064	0.146
	CV [%]	10.88	5.64	2.35	3.94	5.57	7.72	10.9	3.79	1.77	8.50	3.27
Oats	Mean	122^{b}	6.83^{a}	2.54^{b}	3.51^{a}	7.47^{b}	4.22^{a}	1.74^{b}	5.12^{a}	3.55^{b}	1.41^{a}	4.75^{a}
(*n* = 14)	Min	115	6.54	2.40	3.34	7.30	4.10	1.68	4.83	3.47	1.34	4.55
	Max	140	7.21	2.65	3.72	7.77	4.34	1.82	5.46	3.68	1.56	4.97
	SD	6.6	0.181	0.069	0.121	0.129	0.074	0.037	0.150	0.063	0.057	0.113
	CV [%]	5.54	2.66	2.72	3.45	1.73	1.76	2.14	2.93	1.77	4.06	2.38
Rye	Mean	111^{d}	5.06^{b}	2.51^{b}	2.90^{e}	6.16^{d}	3.59^{b}	1.52^{cd}	4.70^{c}	3.23^{d}	1.02^{e}	4.13^{c}
(*n* = 22)	Min	103	4.64	2.42	2.51	5.86	3.29	1.43	4.57	3.13	0.96	3.62
	Max	120	5.27	2.65	3.10	6.32	3.75	1.62	4.84	3.34	1.08	4.29
	SD	4.4	0.147	0.059	0.143	0.104	0.101	0.042	0.069	0.056	0.028	0.152
	CV [%]	3.99	2.90	2.34	4.91	1.68	2.82	2.76	1.48	1.72	2.78	3.69
Triticale	Mean	117^{c}	5.03^{b}	2.45^{c}	3.02^{d}	6.32^{d}	3.23^{d}	1.57^{c}	4.47^{d}	3.05^{e}	1.07^{d}	3.99^{d}
(*n* = 21)	Min	106	4.82	2.34	2.78	6.14	2.98	1.48	4.24	2.90	0.94	3.80
	Max	133	5.32	2.62	3.22	6.67	3.49	1.67	4.68	3.23	1.19	4.18
	SD	6.2	0.149	0.077	0.100	0.127	0.151	0.052	0.106	0.091	0.065	0.117
	CV [%]	5.30	2.97	3.12	3.33	2.02	4.66	3.30	2.38	2.97	6.04	2.93
Wheat	Mean	132^{a}	4.79^{c}	2.53^{b}	3.10^{c}	6.67^{c}	2.72^{f}	1.47^{d}	4.65^{c}	2.86^{f}	1.15^{c}	3.84^{e}
(*n* = 29)	Min	117	4.29	2.44	2.79	6.49	2.41	1.36	4.42	2.74	1.06	3.44
	Max	157	5.20	2.67	3.38	6.90	2.92	1.59	4.93	3.02	1.29	4.16
	SD	9.6	0.226	0.067	0.137	0.121	0.125	0.054	0.109	0.065	0.049	0.185
	CV [%]	7.28	4.72	2.24	4.43	1.82	4.60	3.66	2.35	2.27	4.28	4.82

Notes: ‡SD, standard deviation; ¶CV, coefficient of variation; $^{a–f}$ Means within a column not showing a common superscript letter are significantly different between grain types; All corresponding individual values for each genotype are available online (Tables S4a–4f).

of triticale protein was higher in these feed tables (3.5 and 3.6 vs. 3.2 g Lys/16 g N in the present study). Furthermore, Agroscope (c2011–2015) reported lower Thr concentrations of oat protein (3.3 vs. 3.6 g Thr/16 g N) and the DLG feed table a higher value for wheat protein (3.2 vs. 2.9 g Thr/16 g N). As shown in Table 6, distinct differences existed also in the concentration of other essential AA between the grain proteins. Oat protein was specifically rich in branched-chain AA and Arg, and protein from maize was very high in Leu concentration. Significant differences were also detected in the concentration of several non-essential AA (Table 7). For example, the Ala concentration was the highest in maize protein and the lowest in wheat protein, while Pro was the highest in barley protein and the lowest in oat protein.

Variation in the AA profile of cereal proteins indicates differences in the proportion of individual proteins. Prolamins are rich in Pro and Glu but poor in Lys, whereas albumins and globulins contain less Pro and Glu, but contain more Lys (Draper 1973; Shewry 2007; Klose and Arendt 2012). In support of these results, negative correlations between the concentrations of Lys and Glu as well as Lys and Pro were observed for maize (r = –0.78, r = –0.50), rye (r = –0.52, r = –0.72), triticale (r = –0.51, r = –0.57) and wheat (r = –0.80,

Table 7. Concentration of non-essential amino acids in crude protein of cereal grains.

		Ala	Asp	Cys	Glu	Gly	Pro	Ser	Tyr
		[g/16 g N]							
Barley	Mean	3.92[d]	5.78[e]	2.09[c]	24.3[c]	3.85[d]	12.7[a]	4.39[d]	2.82[c]
(n = 21)	Min	3.68	5.36	1.85	22.5	3.50	11.4	4.25	2.67
	Max	4.17	6.29	2.25	25.7	4.15	13.8	4.61	3.00
	SD[†]	0.151	0.254	0.107	0.85	0.211	0.57	0.100	0.089
	CV[¶] [%]	3.85	4.40	5.11	3.51	5.47	4.49	2.29	3.17
Maize	Mean	7.89[a]	6.70[c]	2.23[b]	18.6[e]	3.71[e]	10.5[d]	5.07[a]	3.70[a]
(n = 27)	Min	7.17	6.30	2.02	17.2	3.27	9.48	4.84	3.43
	Max	8.42	7.09	2.43	19.8	4.20	11.1	5.24	3.83
	SD	0.261	0.203	0.106	0.61	0.249	0.41	0.104	0.108
	CV [%]	3.31	3.03	4.74	3.28	6.71	3.87	2.05	2.91
Oats	Mean	4.85[b]	8.29[a]	2.93[a]	19.9[d]	4.96[a]	5.96[e]	5.07[a]	3.30[b]
(n = 14)	Min	4.69	7.88	2.76	18.4	4.88	5.59	4.91	3.18
	Max	5.03	8.74	3.24	21.1	5.05	6.19	5.22	3.43
	SD	0.080	0.242	0.139	0.67	0.060	0.143	0.090	0.074
	CV [%]	1.65	2.92	4.74	3.38	1.22	2.39	1.77	2.24
Rye	Mean	4.04[c]	6.94[b]	2.10[c]	24.0[c]	4.25[b]	11.5[b]	4.63[c]	2.30[f]
(n = 22)	Min	3.82	6.48	1.94	23.4	4.05	11.1	4.54	2.19
	Max	4.21	7.32	2.21	24.8	4.39	12.2	4.78	2.39
	SD	0.095	0.211	0.077	0.40	0.081	0.33	0.061	0.052
	CV [%]	2.36	3.05	3.66	1.65	1.90	2.85	1.31	2.25
Triticale	Mean	3.77[e]	6.20[d]	2.20[b]	25.8[b]	4.11[c]	10.9[c]	4.68[c]	2.50[e]
(n = 21)	Min	3.57	5.70	2.08	24.4	3.80	10.1	4.51	2.33
	Max	4.04	7.13	2.40	27.2	4.62	11.3	4.98	2.67
	SD	0.122	0.373	0.091	0.67	0.196	0.32	0.111	0.090
	CV [%]	3.23	6.02	4.14	2.58	4.77	2.93	2.37	3.61
Wheat	Mean	3.44[f]	4.99[f]	2.21[b]	29.5[a]	4.04[c]	11.5[b]	4.87[b]	2.67[d]
(n = 29)	Min	3.18	4.59	2.05	28.2	3.87	10.8	4.67	2.54
	Max	3.68	5.34	2.33	31.6	4.35	12.1	5.00	2.84
	SD	0.109	0.202	0.075	0.92	0.121	0.36	0.085	0.075
	CV [%]	3.17	4.04	3.42	3.11	2.99	3.16	1.76	2.81

Notes: [†]SD, standard deviation; [¶]CV, coefficient of variation; [a–f] Means within a column not showing common superscript letter are significantly different between grain types; All corresponding individual values for each genotype are available online (Tables S5a–5f).

$r = -0.47$) ($p \le 0.05$), whereas the concentrations of Pro and Glu were positively correlated in all cereal proteins (barley, $r = 0.90$; maize, $r = 0.56$; oats, $r = 0.54$; rye, $r = 0.82$; triticale, $r = 0.79$; wheat, $r = 0.73$) ($p \le 0.05$) (Table S11).

Compared to protein-rich feed ingredients such as oilseed meals or legume grains, cereals contain only moderate amounts of AA. However, again, because cereals are major dietary ingredients, they represent the largest contributors to dietary AA supply for livestock. Therefore, the variation in AA concentration, observed for some of the cereal grains, is significant for diet formulation. Further studies with animals are warranted to investigate if the observed variations in AA concentration are also reflected in the digestible AA content of the respective cereals.

3.3. *Minerals, inositol phosphates and phytase activity*

3.3.1. *Calcium*

The mean Ca concentration ranged from 0.04 g Ca/kg DM in maize to 1.08 g Ca/kg DM in oats and differed between all cereal grains except for rye and triticale (Table 8). The very low

Table 8. Concentration of different minerals in cereal grains.

		Ca	Mg	K	Na	Fe	Mn	Zn	Cu
		[g/kg DM]			[mg/kg DM]				
Barley	Mean	0.59^{b}	1.63^{a}	5.53^{a}	49.5^{a}	44.4^{b}	15.0^{d}	24.2^{ab}	5.01^{a}
(n = 21)	Min	0.44	1.50	4.84	25.7	31.9	12.1	17.9	4.27
	Max	0.77	1.79	6.28	84.2	75.7	18.3	28.9	6.20
	SD[†]	0.072	0.083	0.402	17.86	10.13	1.68	2.97	0.466
	CV[¶] [%]	12.3	5.08	7.26	36.1	22.8	32.3	12.3	9.29
Maize	Mean	0.04^{e}	1.45^{b}	3.96^{d}	3.40$^{•e}$	22.4^{d}	5.34^{e}	21.3^{cd}	2.04^{d}
(n = 27)	Min	0.03	1.15	3.34	Δ	16.2	3.31	15.6	1.04
	Max	0.06	1.87	5.00	Δ	32.3	10.2	34.0	4.11
	SD	0.009	0.198	0.453	Δ	3.78	1.335	3.67	0.625
	CV [%]	20.0	13.6	11.4	Δ	16.9	25.0	17.2	30.7
Oats	Mean	1.08^{a}	1.45^{b}	3.77^{d}	11.8^{d}	69.1^{a}	29.2^{b}	20.0^{d}	3.64^{c}
(n = 14)	Min	0.95	1.36	3.47	8.11	55.8	22.5	17.0	3.15
	Max	1.33	1.60	4.03	18.4	97.8	33.3	25.7	4.25
	SD	0.096	0.061	0.207	3.05	10.69	3.37	2.51	0.348
	CV [%]	8.93	4.19	5.49	25.8	15.5	11.5	12.6	9.57
Rye	Mean	0.49^{c}	1.36^{c}	5.13^{b}	23.4^{c}	29.8^{c}	19.7^{c}	24.0^{ab}	4.26^{b}
(n = 22)	Min	0.43	1.19	4.47	3.40$^{•}$	23.1	14.1	19.8	3.74
	Max	0.56	1.53	6.16	33.4	40.2	23.7	30.3	4.87
	SD	0.038	0.096	0.418	10.2	4.23	2.46	3.23	0.310
	CV [%]	7.76	7.03	8.16	43.7	14.2	12.5	13.5	7.27
Triticale	Mean	0.49^{c}	1.64^{a}	5.03^{b}	34.0^{b}	31.5^{c}	29.8^{b}	24.4^{a}	4.94^{a}
(n = 21)	Min	0.34	1.40	4.40	3.40$^{•}$	24.2	23.8	18.1	4.28
	Max	0.73	1.92	5.55	54.1	41.5	38.3	30.3	6.32
	SD	0.089	0.143	0.323	16.3	4.49	4.09	2.74	0.490
	CV [%]	18.2	8.73	6.41	48.0	14.3	13.7	11.2	9.92
Wheat	Mean	0.40^{d}	1.56^{a}	4.33^{c}	5.17^{e}	40.9^{b}	32.1^{a}	22.4^{bc}	4.27^{b}
(n = 29)	Min	0.29	1.17	3.78	3.40$^{•}$	31.0	26.2	16.2	3.59
	Max	0.52	2.06	5.18	8.60	54.1	45.4	25.9	5.42
	SD	0.051	0.172	0.383	1.336	5.70	3.71	2.33	0.359
	CV [%]	12.8	11.1	8.83	25.8	13.9	11.6	10.4	8.42

Notes: [†]SD, standard deviation; [¶]CV, coefficient of variation; [•]Mean between limit of detection and limit of quantification; [Δ]Other statistical values not determined, more than 50% of analysed values were below the limit of quantification. [a–e] Means within a column not showing common superscript letter are significantly different between grain types; All corresponding individual values for each genotype are available online (Tables S6a–6f).

level of Ca in maize, which was confirmed by repeated analysis, agreed with the value given in Agroscope (c2011–2015) (0.04 g Ca/kg DM). Vyn and Tollenaar (1998) found even lower values, whereas in other studies a Ca concentration in the range of 0.24–0.66 g Ca/kg DM was reported (Jood et al. 1992; Ullah et al. 2010; Ferreira et al. 2012). It is not clear whether these differences in the reported values can be attributed to differences in analytical methods or other influencing factors, such as location. Ferreira et al. (2012) found differences in the concentration of Ca and other minerals in maize grown on the same location between two years and speculated that this could have resulted from differences in rainfall. In contrast to maize, the Ca concentration of oats in the present study was considerably higher compared to values reported by NRC (2012) (0.33 g Ca/kg DM) and Agroscope (c2011–2015) (0.78 g Ca/kg DM).

3.3.2. *Phosphorus and inositol phosphates*

The mean P concentration ranged from 3.2 g P/kg DM in maize to 4.3 g P/kg DM in barley (Table 9). Means were not significantly different between rye and wheat or

Table 9. Concentration of phosphorus, inositol phosphate phosphorus and phytase activity in cereal grains.

		P [g/kg DM]	Ins(1,5,6) P_3-P [mg/kg DM]	Ins (1,2,3,4,6) P_5-P [mg/kg DM]	Ins (1,2,3,4,5) P_5-P [mg/kg DM]	Ins (1,2,4,5,6) P_5-P [mg/kg DM]	$InsP_6$-P [g/kg DM]	Phytase activity [U/kg DM]
Barley	Mean	4.30^{a}	144^{a}	20.6^{a}	35.0^{a}	29.7^{a}	2.81^{a}	693^{d}
(*n* = 21)	Min	3.91	75.0	11.6	17.8	16.6	2.17	490
	Max	4.73	266	43.5	65.8	65.1	3.52	1100
	SD†	0.264	48.0	10.29	10.70	10.67	0.355	159.8
	CV¶ [%]	5.97	33.3	50.0	30.6	36.0	12.6	23.1
Maize	Mean	3.17^{d}	17.4^{b}	n.d.$^{♦}$	n.d.	22.5^{b}	2.26^{b}	143^{e}
(*n* = 27)	Min	2.59	13.9$^{•}$	n.d.	n.d.	11.6$^{•}$	1.86	100
	Max	4.00	49.3	n.d.	n.d.	40.0	3.09	190
	SD	0.387	$^{\Delta}$	n.d.	n.d.	8.70	0.315	26.4
	CV [%]	12.2	$^{\Delta}$	n.d.	n.d.	38.8	13.9	18.4
Oats	Mean	3.95^{b}	n.d.	n.d.	n.d.	13.8^{c}	1.82^{c}	n.d.
(*n* = 14)	Min	3.59	n.d.	n.d.	n.d.	11.6$^{•}$	1.64	n.d.
	Max	4.45	n.d.	n.d.	n.d.	27.1	1.96	n.d.
	SD	0.218	n.d.	n.d.	n.d.	$^{\Delta}$	0.097	n.d.
	CV [%]	5.52	n.d.	n.d.	n.d.	$^{\Delta}$	5.33	n.d.
Rye	Mean	3.62^{c}	14.9^{b}	12.2^{b}	20.6^{b}	17.1^{c}	1.52^{d}	4177^{a}
(*n* = 22)	Min	3.34	13.9$^{•}$	11.6$^{•}$	14.7	11.6$^{•}$	1.23	3570
	Max	3.82	22.4	17.9	29.0	31.8	1.91	4760
	SD	0.120	$^{\Delta}$	$^{\Delta}$	3.87	5.82	0.175	302.1
	CV [%]	3.32	$^{\Delta}$	$^{\Delta}$	18.8	34.1	11.5	7.23
Triticale	Mean	3.97^{b}	n.d.	15.6^{b}	19.5^{b}	15.0^{c}	1.86^{c}	2154^{b}
(*n* = 21)	Min	3.59	n.d.	11.6$^{•}$	15.5	11.6$^{•}$	1.57	1640
	Max	4.35	n.d.	28.7	24.6	28.6	2.54	2630
	SD	0.227	n.d.	$^{\Delta}$	3.00	$^{\Delta}$	0.200	299.6
	CV [%]	5.71	n.d.	$^{\Delta}$	15.4	$^{\Delta}$	10.8	13.9
Wheat	Mean	3.67^{c}	n.d.	12.1^{b}	18.4^{b}	17.7^{c}	1.92^{c}	1850^{c}
(*n* = 29)	Min	3.24	n.d.	11.6$^{•}$	11.6$^{•}$	11.6$^{•}$	1.38	1340
	Max	4.43	n.d.	16.3	27.1	33.1	2.29	2640
	SD	0.251	n.d.	$^{\Delta}$	4.91	6.22	0.187	295.2
	CV [%]	6.82	n.d.	$^{\Delta}$	26.8	35.1	9.74	16.0

Notes: †SD, standard deviation; ¶CV, coefficient of variation; $^{♦}$n.d., below the limit of detection; $^{\Delta}$Other statistical values were not determined, more than 50% of analysed values were below the limit of quantification; $^{•}$Mean between limit of detection and limit of quantification. $^{a–e}$Means within a column not showing common superscript letter are significantly different between grain types; All corresponding individual values for each genotype are available online (Tables S7a–7f).

triticale and oats. Among grains studied, maize was reported to have the lowest P content (NRC 2012, DLG c2006–2010, Agroscope c2011–2015). It is interesting to note that the proportion of $InsP_6$-P as a percentage of total P was not a constant but varied between 42% in rye and 71% in maize. Maize also showed the highest CV for both P and $InsP_6$-P content and a significant correlation between $InsP_6$-P and EE ($r = 0.46$; $p \leq 0.05$). In the maize kernel, $InsP_6$ is largely associated with the germ. Large variations in EE concentration in the present study were the result of inclusion of both conventional and specialty maize bred for high oil content. Likewise, differences in germ size due to variation in oil content, therefore, also led to different $InsP_6$ concentrations. Significant correlations between $InsP_6$-P and other minerals occurred only in few cases (Table S12).

Concentrations of $InsP_5$ isomers and other $InsP_x$ were very low, if detectable at all. However, for certain isomers, differences were detected between the grain types. All cereal grains contained Ins(1,2,4,5,6)P_5, whereas Ins(1,2,3,4,5)P_5 could only be determined in barley, rye, triticale and wheat. Ins(1,2,3,4,6)P_5 was only present in rye, triticale and wheat. Concerning the $InsP_3$ isomers, only the Ins(1,5,6)P_3 isomer was detected and only in some genotypes of barley, maize and rye. The highest concentrations of $InsP_5$ isomers and $InsP_3$ were found in barley. $InsP_4$ isomers were not detected in any of the grains.

3.3.3. *Phytase activity*

The mean phytase activity was the highest in rye with 4177 U/kg DM, whereas activity in oats was below the limit of detection (100 U/kg). For all cereal grains except oats, variation in phytase activity between genotypes was high and the CV ranged from 7.2% in rye to 23% in barley. Phytase activity in the cereal grains of the present data pool were lower compared to corresponding values reported by Greiner and Egli (2003) and Steiner et al. (2007). Eeckhout and De Paepe (1994), Selle et al. (2003), Shen et al. (2005) and Viveros et al. (2000), however, generally reported similar or lower results compared to the present study. The large variation in reported phytase activity may amongst others be due to different analytical methods, which may strongly influence the results obtained (Greiner and Egli 2003). Furthermore, comparisons of phytase activities may be hampered by comparing miscellaneous cultivars (Steiner et al. 2007) grown in different locations and using different cultivation techniques.

3.3.4. *Other minerals*

Significant differences among grain types were also seen for the other minerals studied (Table 8). The mean Mg content was higher in barley (1.63 g/kg DM), triticale (1.64 g/kg DM) and wheat (1.56 g/kg DM) than in rye (1.36 g/kg DM), oats and maize (both 1.45 g/kg DM) ($p \leq 0.05$). The CV of Mg concentration within grains was lower than for all other minerals displayed in Table 8. The mean concentration of K was the lowest in maize and oats with 4.0 and 3.8 g K/kg DM and the highest in barley with 5.5 g K/kg DM ($p \leq 0.05$). The mean Na content also differed between all cereal grains ($p \leq 0.05$), and was the highest in barley with 50 mg Na/kg DM. The Na content in maize was between the limit of detection and the limit of quantification for all maize genotypes. As shown by the calculated CV, variation in Na content between genotypes of one grain was very high which may be related to analytical inaccuracy because, Na concentrations were close to or below the limit of quantification in many samples.

The mean Fe concentration ranged from 22 mg/kg DM in maize to 69 mg/kg DM in oats, and differed between most of the grains ($p \leq 0.05$). The variation in Fe concentration was the highest in barley and similar for the other grains. The mean Mn concentrations also were significantly different ($p \leq 0.05$) between most of the grains and ranged from 5.3 mg Mn/kg DM in maize to 32 mg Mn/kg DM in wheat. Variation in Mn concentration again was the highest in barley, followed by maize. Differences among grains with regard to the mean Zn concentration and compared to other minerals were relatively low. The mean Zn concentration ranged from 20 mg/kg DM in oats to 24 mg/kg DM in triticale. Regarding Cu, the most obvious finding was the mean concentration was the lowest (2.0 mg/kg DM) and the CV the highest (31%) for maize. All samples were also analysed for their As, Cd and Pb content. However, contents of As, Cd and Pb were below the limit of determination in almost all genotypes. The limit of determination was 0.04 mg/kg for As and 0.025 mg/kg for Cd and Pb.

The variation in Ca, P, Fe, Mn and Zn concentrations observed in the present study may be of significance for livestock feeding, as their concentrations in individual genotypes were close to or below the animal's requirement. In contrast, the large variation in Mg and K between genotypes of some of the cereal grains studied may be less relevant, because they were far above the animal's requirement for all cereal grains and genotypes considered. It should be noted, however, that all grains used in this study (except maize) were grown using the same soil, fertiliser and management conditions. This means that the variations observed here were not the result of fertilisation and soil conditions (Murphy et al. 2009; Spiegel et al. 2009).

3.4. *Thousand seed weight, test weight, falling number and extract viscoelasticity*

3.4.1. *Thousand seed weight, test weight and falling number*

The mean TSW was 288 g/1000 seeds in maize and ranged from 39 to 59 g/1000 seeds in the soft grains (Table 10). Differences in TSW were significant ($p \leq 0.05$) except for

Table 10. Physical characteristics of the grains.

		Thousand seed weight [g/1000 seeds]	Test weight [kg/hl]	Falling number [s]
Barley	Mean	58.8^{b}	71.7^{c}	368^{a}
(*n* = 21)	Min	52.3	66.5	255
	Max	66.0	73.9	443
	SD†	3.76	1.99	48.8
	CV¶ [%]	6.38	2.77	13.3
Maize	Mean	288^{a}	75.4^{b}	–
(*n* = 27)	Min	219	67.6	–
	Max	340	80.6	–
	SD	27.8	3.23	–
	CV [%]	99.63	4.28	–
Oats	Mean	38.5^{d}	54.7^{d}	99.1^{c}
(*n* = 14)	Min	31.3	51.3	62.0
	Max	46.4	57.6	227
	SD	4.50	2.03	46.9
	CV [%]	11.7	3.70	47.3
Rye	Mean	41.5^{d}	76.5^{b}	181^{b}
(*n* = 22)	Min	38.1	71.0	87.0
	Max	44.3	79.5	336
	SD	1.89	2.36	58.8
	CV [%]	4.55	3.09	32.5
Triticale	Mean	50.3^{c}	75.3^{b}	99.6^{c}
(*n* = 21)	Min	43.8	70.2	62.0
	Max	61.4	78.6	293
	SD	4.30	1.90	55.5
	CV [%]	8.55	2.53	55.8
Wheat	Mean	51.5^{bc}	81.1^{a}	344^{a}
(*n* = 29)	Min	44.9	77.7	229
	Max	59.7	85.1	402
	SD	3.54	1.73	48.4
	CV [%]	6.87	2.14	14.01

Notes: [†]SD, standard deviation; [¶]CV, coefficient of variation; -, not determined; [a–d] Means within a column not showing common superscript letter are significantly different between grain types; All corresponding individual values for each genotype are available online (Tables S8a–8f).

triticale and wheat, and for barley and wheat. These results are in close agreement with previous reports (Metayer et al. 1993; Ullah et al. 2010). The variation between genotypes was the highest in oats (CV 12%) and lower in the other grains. The mean TW was the highest in wheat (82 kg/hl) and the lowest in oats (55 kg/hl). Variability of TW between genotypes within grain type was relatively low (CV 2.1–4.3%). Previous reports showed lower TW values (Metayer et al. 1993; Svihus and Gullord 2002), but similar values for barley (67 kg/hl), maize (76 kg/hl), oats (56 kg/hl) and triticale (74 kg/hl) were recorded by Agroscope (c2011–2015). Differences in TW among cereal grains apparently reflect differences in bulk density. Naked cereal varieties and genotypes having a higher bulk density, such as wheat and rye, had higher TW values than hulled cereals, such as oats and barley (Andersson et al. 1999). The mean FN showed very large differences between grain types and genotypes within grain type. It ranged from 99 s in oats to 368 s in barley.

As chemical analysis of feed ingredients is time consuming, labour intensive and expensive, the industry could benefit from a method for the rapid prediction of nutritional composition of cereal grains, e.g. based on physical properties. We found several significant correlations between physical traits and nutrient fractions. For example, in barley, TSW was positively correlated with CP concentration (r = 0.48; $p \leq 0.05$) and negatively correlated with CF concentration (r = −0.50; $p \leq 0.05$) and NDF concentration (r = −0.45; $p \leq 0.05$). However, the coefficients of correlation although statistically significant generally were not high enough to use them as predictor for a specific nutrient concentration.

3.4.2. *Extract viscoelasticity*

When applied in animal nutrition research, rheological properties of cereals usually are determined as the apparent extract viscosity, which is measured at one selected shear rate. Viscosity values measured at different shear rates fitted to the Herschel–Bulkley model enable to describe the viscoelastic properties of fluids (Steffe 1996) like cereal extracts by estimating the yield point, consistency index and flow index. The average yield point ranged from −43.4 mPa in rye to 9.81 mPa in maize (Table 11). For all cereal grains, variation for yield point was high, with a range of 5.31 mPa (CV 26.6%) in triticale and 490 mPa (CV incalculable due to negative values) in rye. Yield points represent extrapolated values and cannot be interpreted physically if negative, but yield point estimates are necessary to achieve precise estimates for consistency and flow indices. The average consistency index varied between 0.24 mPa · s^n in maize and 112 mPa · s^n in rye, with a range within cereal grains from 0.21 mPa · s^n (CV 10.9%) in oats to 362 mPa · s^n (CV 77.4%) in rye. The flow index indicates whether the viscosity of a fluid increases or decreases when an increasing shear stress impacts on a fluid, e.g. induced by peristalsis. The average flow index varied between 0.74 in rye and 1.20 in maize, with a range of cereal grains from 0.03 (CV 0.7%) in triticale to 0.20 (CV 7.1%) in rye. Thus, the viscosity of rye decreased, whereas the viscosity of the other cereal grains especially maize increased under increasing shear stress.

At an exemplary shear rate of 380 s^{-1} (medium of reported values in literature), the average extract viscosity was 0.76 mPa · s for maize, 0.95 mPa · s for oats, 1.12 mPa ·s for wheat, 1.26 mPa · s for triticale, 1.94 mPa · s for barley and 20.0 mPa · s for rye. The level of extract viscosity is difficult to compare between studies due to methodological differences during sample preparation. However, the same ranking of apparent extract viscosity values

Table 11. Extract viscoelasticity of the grains.

		τ_0[♦] [mPa]	k[†] [mPa · s^n]	n[§]	Extract viscosity[$] [mPa · s]
Barley	Mean	5.35[a]	1.72[b]	1.03[d]	1.94[b]
(n = 21)	Min	3.21	0.97	0.98	1.27
	Max	9.38	7.65	1.04	6.96
	SD[‡]	1.56	1.42	0.01	1.22
	CV[¶] [%]	29.2	82.6	1.27	63.1
Maize	Mean	9.81[a]	0.235[b]	1.20[a]	0.76[f]
(n = 27)	Min	5.64	0.075	1.17	0.73
	Max	40.21	0.285	1.36	0.79
	SD	6.39	0.040	0.04	0.01
	CV [%]	65.1	18.0	2.95	1.54
Oats	Mean	6.15[a]	0.63[b]	1.07[b]	0.95[e]
(n = 14)	Min	4.41	0.50	1.05	0.91
	Max	10.01	0.73	1.10	0.99
	SD	1.55	0.07	0.01	0.02
	CV [%]	25.2	10.9	1.36	2.40
Rye	Mean	−43.4[b]	112.1[a]	0.74[e]	20.0[a]
(n = 22)	Min	−239.5	28.6	0.62	9.68
	Max	250.2	361.5	0.82	41.0
	SD	94.6	86.8	0.05	7.99
	CV [%]	–	77.4	7.11	39.9
Triticale	Mean	4.53[a]	0.99[b]	1.04[cd]	1.26[c]
(n = 21)	Min	3.08	0.78	1.02	1.06
	Max	8.39	1.49	1.05	1.72
	SD	1.21	0.16	0.01	0.14
	CV [%]	26.6	15.8	0.69	11.3
Wheat	Mean	8.92[a]	0.81[b]	1.06[bc]	1.12[d]
(n = 29)	Min	2.82	0.12	1.03	0.96
	Max	124.20	1.11	1.33	1.39
	SD	22.30	0.19	0.05	1.14
	CV [%]	250	23.3	5.03	12.7

Notes: [♦]τ_0, yield point; [†]k, consistency index; [§]n, flow index (dimensionless); [$]Shear rate of 380 s^{-1}; [‡]SD, standard deviation; [¶]CV, coefficient of variation; [a–f] Means within a column not showing common superscript letter are significantly different between grain types; All corresponding individual values for each genotype are available online (Tables S9a–9f).

between cereal grains was described in the literature for barley and wheat (Grosjean et al. 1999b), and for maize, wheat and triticale (Çiftci et al. 2003). A variation in apparent extract viscosity between wheat genotypes was also reported in the literature (Dusel et al. 1997; Grosjean et al. 1999a, 1999b). We determined no significant correlation between extract viscosity at a shear rate of 380 s^{-1} and aNDFom for any grain type. Extract viscosity at a shear rate of 380 s^{-1} was negatively correlated ($p \le 0.05$) with the fructans and uronic acids concentration ($r = -0.76$ and $r = -0.83$, respectively) in barley (Table S13). It was positively correlated ($p \le 0.05$) with the concentrations of some NSP fractions (soluble arabinose, $r = 0.58$; soluble xylose, $r = 0.62$; total arabinose, $r = 0.82$; total xylose, $r = 0.72$; galactose, $r = 0.54$; glucose, $r = 0.45$; cellulose, $r = 0.46$) in rye (Table S14). It was also positively correlated ($p \le 0.05$) with the total galactose concentration ($r = 0.49$) in wheat. Positive correlations between extract viscosity and soluble pentosan concentrations in wheat were also determined by Dusel et al. (1997). Dusel et al. (1997) further showed that the extract viscosity was influenced by other factors that are affected by different conditions during cultivation. Such other factors may be the average size and structure of the soluble arabinoxylan molecules (Saulnier et al. 2007) and other soluble substances in cereal grains like gliadin and glutenin (Wang et al. 2004). The high level of extract viscosity especially in rye may also be related to certain protein fractions, because Weipert (1997) found rye to have a high content of water-extractable proteins compared to other cereal grains.

4. Conclusion

The present study confirmed that cereal grains of different genotypes substantially differ in their chemical composition and physical characteristics. In some characteristics, average chemical composition as determined herein differed from values reported in common feed tables, while for other characteristics the values were similar. Because the cereals had been grown under well-standardised conditions, effects location and agronomy may have on chemical composition still need to be investigated. Animal studies were conducted to better evaluate the relevance of the detected variability for different animal species. Results from the animal trials are subject of other communications.

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Disclosure statement

No potential conflict of interest was reported by the authors.

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