The nutritional value of new varieties of high-yielding triticale: Nutrient composition and *in vitro* digestibility

A.E. Widodo[1,2], J.V. Nolan[1] & P.A. Iji[1#]

[1]School of Environmental and Rural Science, University of New England,
Armidale, NSW 2351, Australia
[2]Department of Animal Science, University of Papua, Manokwari - West Papua 98314, Indonesia

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Abstract

Ten high-yielding cultivars of triticale obtained from a breeding group at the University of New England, harvested in 2008 and 2009, were assessed for nutrient composition and nutrient digestibility. The cultivars tested were AT528, H20, H127, H128, H157, H249, H418, H426, JRCT74 and Tahara. Their nutrient characteristics, including dry matter, crude protein, crude fat, ash and gross energy, starch content and composition, and concentrations of non-starch polysaccharides, minerals, phytate-P content and amino acids, were determined. The *in vitro* digestibility and viscosity during digestion were also measured. There was low variability between the cultivars tested and harvest years (the difference between wet and dry conditions) had little effect on nutrient composition.

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Introduction

Triticale *(X Triticosecale Wittmack)* is the product of a cross between wheat *(Triticum)* and rye *(Secale).* It has been developed primarily as a feed grain for livestock owing to its low versatility for the human food market compared with other conventional cereal grains such as wheat (King, 2011). The world production of triticale has more than doubled in recent years, from 6.4 million tonnes in 2006 to 13.7 million tonnes in 2009 (FAO, 2010). Although it is a minor cereal grain compared with wheat, barley, sorghum and maize, the domestic use of this cereal for animal feed increased in Australia, for example from 202 200 tonnes in 2006 to 788 000 tonnes in 2010 (To *et al.*, 2010).

Since the first commercial triticale cultivars were released in 1969, many studies have been carried out on the development of triticale. Triticale demonstrates many agronomic advantages, including winter hardiness, drought and disease tolerance and excellent productivity potential. However, they exhibit a wide variation in nutrient content between the cultivars (Vieira *et al.*, 1995).

Over the past 25 years, breeding and selection programmes in Australia have been active and are still being undertaken in New South Wales, South Australia and Western Australia to improve the quality and yield of triticale (Andrews *et al.*, 1991). A crop breeding group at the University of New England (UNE) has developed varieties that are higher yielding and more disease resistant and tolerant of acid soil than the current commercial strains, as well as being high in energy content (UNE, 2008). This group has bred cultivars that have broken the 10-tonne-per-hectare barrier for a wheat-related grain crop. In comparison, Australia's average wheat yield is only 2 tonnes/hectare, while in Europe, wheat yields do not exceed 6 to 8 tonnes/hectare (Scanlan, 2005). These varieties hold much promise for livestock industries, which could use this grain as an alternative energy source. However, there have been no studies on their nutritional composition and value as broiler feed ingredients. This study was therefore conducted to evaluate nutrient composition and *in vitro* digestibility of new triticale cultivars produced by UNE's crop breeding group.

Materials and Methods

Ten triticale cultivars were investigated. They were obtained from UNE's crop breeding group, harvested from the university's experimental plots at Armidale (30.50° S, 151.65° E; altitude, 970 m above sea level) in 2008 and 2009 . According to the breeding group, there were marked differences in harvest

conditions between the two years, 2009 being wetter (816.4 mm) than 2008 (766.8 mm). The experiment was conducted in the UNE's field laboratory in Armidale. The 10 high-yielding triticale cultivars tested were AT528, H20, H127, H128, H157, H249, H418, H426, JRCT74 and Tahara.

About 5 kg of each cultivar was received from the breeders and ground to pass through a 1 mm sieve. Sub-samples were then taken from this bulk for laboratory analyses. For starch granule analysis, a sub-sample of whole grains was collected and stored in 20 mL containers. The analyses of the samples are described below.

The dry matter (DM) content was conducted by the following method. Approximately 2 g were weighed in duplicate into cooled pre-weighed silica crucibles and placed in a forced-air convection oven (Qnaltex Universal Series 2000, Watson Victor Ltd., Perth, Australia), which was preheated to 105 ºC. Samples were set at this temperature for 24 h and then cooled in the desiccator for 40 to 45 min to a constant weight. The DM was calculated based on the water loss in weight of the sample after drying.

The nitrogen (N) content of the samples was determined according to the Dumas combustion technique (AOAC, 2002; ID 990.03) with a LECO[®]FP2000 automatic N analyser (LECO Corporation, St. Joseph, MI, USA). In order to interpret the detector response as percentage N (w/w), calibration was carried out using a primary standard of pure ethylenediaminetetraacetic acid (EDTA). A factor of 5.76 was used to calculate crude protein (CP) from the total N (McKenzie & Farrell, 1980). Crude fat or ether extract (EE) content was determined on a Soxhlet apparatus (AOAC, 2002; ID 920.39). Ash content was determined according to AOAC (2002; ID 942.05) method, and dry samples were ignited in a Carbolite CWF Furnace 1200 chamber furnace (Carbolite, Sheffield, UK). The gross energy (GE) concentration was determined for individual samples, using IKA[®] WERKE bomb calorimeter (C 7000, GMBH & Co., Staufen, Germany). The chamber was calibrated for the heat of combustion of 0.5 g of benzoic acid (26.457 kJ/g) prior to sample analysis. Approximately 0.50 g of samples was weighed in duplicate and combusted. The GE value was directly obtained digitally as MJ/kg.

Total starch and resistant starch in the samples were determined with the Megazyme total starch kit (Megazyme International, Ireland), using the enzyme procedure developed by McCleary *et al.* (1994). The amylose and amylopectin contents were determined with a Megazyme amylose/amylopectin assay kit (Megazyme International Ireland, Bray Business Park, Bray, Ireland) using the selective quantitative precipitation reaction of con-canavalin A (Con A) for amylopectin (Gibson *et al.*, 1997) and by the colorimetric method of iodine binding with amylose (Chrastil, 1987).

Soluble and insoluble non-starch polysaccharides (NSP) of the ground samples were measured as described by Englyst & Hudson (1993) and Theander & Westerlund (1993). The free sugars were determined using the supernatant kept for this purpose, by hydrolysing with 1 mL of 12 M H_2SO_4 at 30 ºC for 2 h (Saeman *et al.*, 1963), then reducing and acetylating the sugar as per the previous two analyses. The released sugars in all three solutions were measured on a gas chromatograph (Hewlett Packard Model 427, Packard Instruments, Sydney, NSW, Australia) after calibration with allose as internal standard.

Quantitative amino acid (AA) analysis of the ground samples of triticale cultivars was conducted at the Australian Proteome Analysis Facility Ltd, Macquarie University. About 100 mg of sample underwent liquid hydrolysis in 6 M HCl at 110 ºC for 24 h. After hydrolysis, all amino acids except for cysteine and tryptophan were analysed using the Waters AccQTag Ultra Chemistry (Rydalmere NSW, Australia). The samples were analysed in duplicate. Cysteine and tryptophan were not determined in this study because of the method used.

The mineral contents of the ground grain samples were determined by the microwave digestion technique (Yang, *et al.*, 2012). The samples, 0.45 g, were weighed into a Teflon® TFM vessel. In a fume hood, 8 mL of nitric acid (70% v/v) was added along with 2 mL of hydrogen peroxide (30% v/v). The solution was swirled to homogenise it. The vessel was closed and introduced to the rotor segment, then tightened, using a torque wrench. The segment was inserted into the microwave cavity and the temperature sensor connected. The microwave programme was run to completion (about 45 min) and the rotor cooled by air until the solution reached room temperature. The vessel was opened and the solution quantitatively transferred into a 50-mL volumetric flask. The solution was made to 50 mL total volume with deionised water and mixed well prior to analysis. The subsequent mineral determination was carried out by radial system Varian model VISTA-MPX Simultaneous, Inductively Coupled Plasma-Optical Emission Spectrometer (Varian[©] Australia Pty Ltd). The phytate-P content of the grain samples was determined using a sensitive method for the rapid determination of phytate-P in cereals and cereal product as described by Haug & Lantzsch (1983).

In vitro digestibility was carried out according to the method described by Babinszky *et al.* (1990), with slight modifications. Digestibility was measured for DM, CP and starch. Five hundred mg of ground samples were digested in triplicate in 12.5 mL of 0.1 M HCl containing 4 g pepsin/L (Sigma Chemical, St. Louis. Mo, USA) and the sample was incubated at 40 ºC for 1.5 h. Then the sample was dissolved in 2 mL of 110 mg $NaHCO_3$ and 12.5 mL of potassium phosphate buffer (pH 6.8) containing 4 g of pancreatin and 4 mL of

amylase per litre. The mixture was incubated at 40 ºC for 3 h. After the incubation period, 2.5 mL of $NaCO_3$ (100 g/L) were added to each tube and the contents were centrifuged at 3500 × *g* for 15 min. The supernatant was collected and kept on ice until the viscosity analysis and the residue was repeatedly rinsed with Milli-Q water. The residue then was freeze-dried and used for determination of DM, CP and starch. Starch was calculated by converting from glucose content. The digestibility of DM, CP and starch was calculated by the following equation:

Digestibility (%) =
$$\frac{\text{Weight of samples or nutrient (mg)} - \text{Weight of dried residue or residual nutrient (mg)}}{\text{Weight of DM or nutrient in samples (mg)}} \times 100$$

The supernatant collected from the *in vitro* analysis was used to measure viscosity of the grain as described by Bedford & Classen (1993). The Brookfield DVIII viscometer was used, which was set at 25 ºC with a cP 40 spindle at 100 rpm. The values were expressed in centipoise (cP), which is 1/100 dyne second per cm^2.

Data were analysed using 2-sample t-test (α <0.05) in Minitab 16 (Minitab, 2010) to determine whether or not the nutrients of the cultivars were different between the two different harvest years. If the *P*-value was less than or equal to the α level, the null hypothesis (H_0) was rejected and it was concluded that there were significant differences in the nutritional value of the cultivars harvested in 2008 and 2009. The variation in nutrients within the harvest year was evaluated as coefficient of variation (CV). A correlation analysis was conducted to determine the relationship between the parameters.

Results

The proximate composition of the cultivars in the two harvest years is shown in Table 1. The DM content varied from 865.5 (H127; harvested in 2009) to 882.7 g/kg (H418; 2009) and the CP content ranged between 92.0 (H128; 2009) and 127.8 g/kg (H418; 2008). The crude fat content varied from 13.9 (H128; 2008) to 27.4 g/kg (Tahara; 2008), while ash concentration was from 16.7 (Tahara; 2008) to 20.0 g/kg (H157; 2009) and GE values were between 18.1 (H128; 2009) and 18.5 MJ/kg (Tahara; 2008). The DM, CP, ash content and GE were more uniform (*P* >0.05; with CV <10%), regardless of harvest year, while EE (CV >15%) was more variable.

The starch content ranged from 577.6 (H157; 2008) to 661.6 g/kg (H127; 2008) (Table 2). The values of amylose ranged from 137.9 (Tahara; 2009) to 180.4 g/kg (H249; 2008), whereas the amylopectin content varied from 429.1 (H157; 2008) to 499.8 g/kg (H128; 2009). In percentage terms, amylose was 23.2% - 27.8%, while amylopectin varied between 72.2% and 76.8%. The resistant starch values were between 41.8 (JRCT; 2008) and 50.9 g/kg (H128; 2009). In addition, the amylose : amylopectin ratio ranged from 0.30 (Tahara; 2009) to 0.38 (H20; 2009 and H249; 2008). The total starch, amylose, amylopectin, amylose : amylopectin ratio and resistant starch contents were fairly uniform (CV between 3.65% and 8.47%) and were not different (*P* >0.05) between the harvest years.

The total free sugar values varied from 19.74 (H157; 2009) to 32.75 g/kg DM (H426; 2008) (Table 3). Total soluble NSP content ranged between 8.34 (H157; 2009) and 15.09 g/kg DM (AT528; 2008) while insoluble NSP was between 79.16 (H20; 2009) and 122.06 g/kg (Tahara; 2009) (Tables 4 and 5, respectively). The concentrations of the individual free sugars, soluble and insoluble NSP were uniform (*P* >0.05) between the two harvest years. However, the galactose content of free sugars and mannose component of soluble NSP were different (*P* <0.05) between cultivars harvested in 2008 and 2009.

The AA contents of the triticale cultivars are shown in Table 6. The cultivars were richest in glutamine and proline. The major essential amino acids in the triticale cultivars were arginine, glycine, valine, leucine and phenylalanine. There were no differences (*P* >0.05) in amino acid concentrations between the cultivars between the two harvest years as shown by the low CV (from 3.47% to 12.93%).

Considerable variation was observed for the mineral contents of triticale cultivars across the two harvest years as shown in Table 7. Calcium content ranged from 0.31 (H128; 2009, H418; 2009 and H426; in 2008) to 0.46 g/kg (Tahara; 2008), while the P content varied from 3.50 (H418; 2009) to 4.42 g/kg (H157; 2008). The values for K content were between 4.0 (H418; 2009) and 6.1 g/kg (H157; 2009). Sulphur content varied from 1.2 (H128; 2009) to 1.7 g/kg (H249; 2009) and the Mg content ranged from 1.4 (H127; 2009) to 1.9 g/kg (H418; 2008). The concentrations of Ca, K, S and Mg were similar (*P* >0.05, CV <30%) between the two harvest years; however, P content was different (*P* = 0.07) between 2008 and 2009, but it had a low

Table 1 Proximate composition (g/kg DM) of 10 triticale cultivars in two harvest years

Item	AT528		H20		H127		H128		H157		H249		H418		H426		JRCT74		Tahara		CV%[1]		P
	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	
Dry matter	873.4	874.9	872.6	872.2	873.4	865.5	873.1	876.3	873.3	875.6	873.6	874.9	879.1	882.7	871.2	877.9	873.2	879.1	873.1	878.0	0.24	0.52	0.210
Crude protein	115.4	106.0	120.8	124.2	115.4	124.4	124.7	92.0	125.3	115.1	118.0	125.1	127.8	114.2	121.7	118.8	122.7	127.8	116.1	108.2	3.67	9.58	0.195
Crude fat	15.9	20.6	18.4	16.3	15.9	18.8	13.9	20.4	14.6	19.3	13.9	17.5	18.5	17.3	19.1	26.7	19.8	18.5	27.4	23.6	22.72	15.83	0.202
Ash	17.4	18.5	19.8	18.1	19.2	18.3	18.1	19.8	18.7	20.0	17.2	17.5	17.4	17.9	17.5	18.1	19.9	19.2	16.7	18.49	6.22	4.45	0.396
Gross energy[2]	18.2	18.2	18.3	18.4	18.2	18.2	18.2	18.1	18.4	18.3	18.3	18.4	18.4	18.3	18.4	18.3	18.4	18.4	18.5	18.3	0.50	0.55	0.371

[1] CV: coefficient of variation.
[2] MJ/kg DM.

Table 2 Total starch, amylose, amylopectin, resistant starch contents (g/kg DM) and amylose : amylopectin ratio of 10 triticale cultivars in two harvest years

Item	AT528		H20		H127		H128		H157		H249		H418		H426		JRCT74		Tahara		CV%[1]		P
	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	
Total starch	653.7	634.6	622.3	612.3	661.6	653.7	638.9	657.0	577.6	613.6	652.5	606.8	592.3	612.3	626.1	590.0	604.0	612.3	592.3	595.4	4.75	3.65	0.780
Amylose	171.2	155.5	162.1	170.2	168.4	170.1	169.7	157.3	148.5	161.2	180.4	163.1	144.8	159.8	146.5	146.6	150.1	148.1	142.9	137.9	8.47	6.59	0.786
Amylopectin	482.5	479.1	460.2	442.1	493.2	483.5	469.2	499.8	429.1	452.3	472.1	443.7	447.5	452.5	479.6	443.4	453.9	464.2	449.4	457.5	4.16	4.25	0.833
Resistant starch	47.8	48.0	46.1	45.5	48.4	47.2	48.4	50.9	46.3	44.1	49.7	43.8	44.5	45.7	45.3	44.1	41.8	44.2	45.3	47.1	4.97	4.91	0.772
Am : Ap	0.35	0.32	0.35	0.38	0.34	0.35	0.36	0.31	0.35	0.36	0.38	0.37	0.32	0.35	0.31	0.33	0.33	0.32	0.32	0.30	6.66	7.75	0.916

[1] CV: coefficient of variation; Am : Ap = amylose : amylopectin ratio.

Table 3 Available free sugar contents (g/kg DM) of 10 triticale cultivars in two harvest years

Item	AT528		H20		H127		H128		H157		H249		H418		H426		JRCT74		Tahara		CV%[1]		P
	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	
Arabinose	0.57	0.47	0.38	0.43	0.58	0.49	0.43	0.61	0.37	0.38	0.43	0.36	0.58	0.45	0.44	0.51	0.46	0.55	0.42	0.53	17.47	15.86	0.759
Xylose	0.21	0.00	0.15	0.19	0.28	0.21	0.22	0.25	0.17	0.00	0.19	0.15	0.29	0.00	0.21	0.19	0.00	0.00	0.00	0.19	58.34	88.59	0.256
Mannose	4.3	4.1	5.7	4.8	3.7	3.0	4.2	3.3	3.6	3.3	4.0	4.1	4.9	2.7	4.7	5.5	4.1	5.3	3.4	4.8	15.66	24.24	0.639
Galactose	1.3	2.0	1.7	2.3	1.5	1.7	1.3	1.9	1.3	1.9	1.5	2.6	1.7	1.8	1.6	2.3	1.2	1.7	1.9	2.3	15.07	14.95	0.000
Glucose	16.6	17.8	18.5	23.2	21.2	15.1	23.1	15.8	17.9	14.2	14.3	20.0	22.9	14.9	25.6	22.2	17.9	20.6	16.5	18.6	18.39	17.59	0.444
Total	23.1	24.4	26.5	31.0	27.2	20.4	29.4	21.7	23.4	19.7	20.5	27.2	30.9	19.9	32.8	30.6	23.6	28.1	22.2	26.4	15.65	17.59	0.591

[1] CV: coefficient of variation.

Table 4 Soluble non-starch polysaccharides contents (g/kg DM) of 10 triticale cultivars in two harvest years

Item	AT528		H20		H127		H128		H157		H249		H418		H426		JRCT74		Tahara		CV%[1]		P
	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	
Arabinose	5.2	5.4	4.3	3.4	4.4	3.9	3.6	4.1	3.2	2.9	3.8	3.0	3.0	3.3	2.9	3.4	3.9	3.8	5.1	4.1	20.78	19.42	0.570
Xylose	6.9	6.9	5.7	4.5	5.8	5.0	5.1	5.4	4.3	4.0	4.7	3.7	4.1	4.5	3.6	4.4	4.7	4.9	6.3	5.1	20.31	18.73	0.500
Mannose	1.2	0.5	1.1	1.2	1.1	0.4	0.9	0.5	1.0	0.4	1.1	1.1	1.2	0.4	1.3	0.4	0.4	0.4	0.4	0.5	32.98	52.55	0.013
Galactose	1.3	1.2	1.3	1.2	1.1	1.1	0.9	1.1	1.2	1.2	1.2	1.3	1.0	1.1	1.2	1.2	1.1	1.0	1.3	1.2	11.64	7.27	1.000
Glucose	2.4	1.8	2.5	2.0	1.6	1.7	1.4	1.8	1.2	1.0	1.8	1.4	1.8	2.1	2.2	1.7	1.5	1.6	2.1	2.0	23.92	19.80	0.436
Total	15.1	14.0	13.3	10.9	12.3	10.7	10.5	11.5	9.7	8.3	11.2	9.2	9.8	10.1	10.0	10.0	10.3	10.5	13.5	11.5	16.22	14.35	0.256

[1] CV: coefficient of variation.

Table 5 Insoluble non-starch polysaccharides contents (g/kg DM) of 10 triticale cultivars in two harvest years

Item	AT528		H20		H127		H128		H157		H249		H418		H426		JRCT74		Tahara		CV%[1]		P
	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	
Arabinose	30.7	31.0	28.8	23.9	27.6	30.6	29.7	30.6	34.2	41.0	30.7	27.0	29.2	38.1	27.7	34.2	35.9	37.8	38.4	43.1	11.70	18.37	0.302
Xylose	42.3	39.1	38.2	30.9	36.6	38.4	36.6	36.9	44.7	50.0	38.2	32.6	36.0	48.3	36.6	43.7	44.0	47.2	48.5	50.1	10.90	17.09	0.567
Mannose	2.8	4.2	3.4	4.7	4.1	4.6	5.1	4.4	4.3	4.3	2.9	4.2	4.0	4.6	5.1	5.5	4.2	4.3	4.8	4.9	20.81	8.76	0.110
Galactose	2.6	2.3	2.6	2.6	2.8	2.4	3.2	2.3	3.7	2.9	2.9	2.8	3.5	3.4	3.2	2.8	3.2	2.9	3.4	3.6	12.63	15.61	0.124
Glucose	33.6	30.4	27.5	27.4	31.7	26.4	26.3	23.9	36.6	33.4	28.5	31.5	31.4	35.1	28.9	28.6	30.4	30.6	33.9	36.0	10.37	12.73	0.738
Total	99.5	95.4	89.4	79.2	91.3	91.1	89.4	87.3	109.5	117.0	91.8	87.0	92.3	114.9	90.2	102.4	104.7	109.1	114.8	122.1	9.58	14.63	0.563

[1] CV: coefficient of variation.

Table 6 Amino acid concentrations (g/kg DM) of triticale harvested in different years

Item	AT528		H20		H127		H128		H157		H249		H418		H426		JRCT74		Tahara		CV%[1]		P
	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	
Alanine	5.7	5.0	5.6	5.8	5.8	5.7	5.1	4.5	5.6	5.1	5.5	6.0	5.6	6.0	5.3	5.6	5.6	6.0	5.4	5.2	3.70	9.22	0.821
Arginine	7.6	6.6	7.5	7.4	7.7	7.1	6.6	5.4	7.6	6.6	7.4	7.9	7.0	7.6	6.9	7.4	7.5	7.5	7.4	7.0	4.89	10.04	0.283
Aspartic acid	8.8	7.5	8.0	8.4	9.0	8.9	7.7	6.3	8.3	7.3	8.2	8.6	7.8	8.5	7.7	8.1	8.2	8.5	8.1	7.6	5.10	9.92	0.491
Glutamic acid	36.6	30.8	35.5	34.9	37.0	37.8	30.3	24.7	34.7	33.1	36.4	36.9	43.4	37.8	35.0	32.9	34.4	37.5	33.2	28.9	9.39	13.10	0.242
Glycine	6.1	5.7	6.3	6.2	6.1	5.8	5.7	4.6	6.2	5.7	6.3	6.4	6.1	6.3	5.7	6.0	6.2	6.3	6.1	5.9	3.56	8.86	0.333
Histidine	3.5	2.9	3.5	3.4	3.5	3.3	2.9	2.5	3.5	3.1	3.4	3.6	3.6	3.4	3.2	3.4	3.3	3.4	3.2	3.1	6.17	10.01	0.265
Isoleucine	4.8	4.2	4.7	4.7	4.9	4.9	4.1	3.6	4.8	4.3	4.8	5.0	5.0	4.9	4.5	4.6	4.6	4.9	4.5	4.2	5.53	9.88	0.406
Leucine	9.3	8.1	9.0	8.9	9.4	9.3	8.1	6.9	9.0	8.4	9.2	9.5	9.8	9.2	8.7	8.7	8.9	9.2	8.8	8.0	5.32	9.12	0.191
Lysine	4.8	4.3	4.7	4.7	4.8	4.7	4.3	3.8	4.7	4.3	4.6	4.9	4.4	4.6	4.4	4.7	4.6	4.6	4.6	4.8	3.77	6.89	0.488
Methionine	1.8	1.7	2.0	2.0	1.8	1.9	1.6	1.4	2.0	1.5	1.9	2.0	1.8	1.9	1.9	1.7	2.0	1.9	1.8	1.8	5.84	2.50	0.173
Phenylalanine	6.6	5.6	6.3	6.0	6.6	6.5	5.6	4.7	6.4	5.7	6.4	6.5	7.1	6.3	6.1	6.0	6.1	6.2	6.1	6.6	6.32	9.49	0.060
Proline	13.3	11.5	13.4	12.7	13.4	13.1	11.3	9.3	13.1	12.2	13.7	13.7	15.5	13.6	13.1	12.1	12.9	13.5	12.0	13.3	8.28	11.93	0.118
Serine	6.5	5.7	6.6	6.3	6.6	6.4	5.7	4.8	6.5	6.0	6.6	6.7	7.1	6.7	6.2	6.2	6.5	6.7	6.5	6.5	5.45	9.42	0.185
Threonine	4.6	3.9	4.4	4.4	4.6	4.5	3.9	3.4	4.4	3.9	4.4	4.7	4.6	4.4	4.3	4.2	4.4	4.4	4.5	4.6	4.71	8.94	0.111
Tyrosine	3.0	2.5	2.8	2.7	3.1	2.5	2.5	1.9	2.7	2.7	2.9	3.0	2.8	2.8	2.8	2.6	2.9	2.8	2.6	3.0	6.38	11.58	0.066
Valine	6.5	5.6	6.4	6.4	6.6	6.4	5.5	4.9	6.4	5.8	6.4	6.8	6.5	6.5	6.1	6.2	6.3	6.5	6.1	5.7	5.08	9.39	0.350

[1] CV: coefficient of variation.

Table 7 Concentrations of macro and trace minerals of triticale cultivars (DM) harvested in different years

Item		AT528		H20		H127		H128		H157		H249		H418		H426		JRCT74		Tahara		CV%[1]		P
		2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	
Ca		0.4	0.3	0.4	0.4	0.4	0.3	0.4	0.3	0.4	0.4	0.4	0.4	0.4	0.3	0.3	0.3	0.4	0.4	0.5	0.4	11.79	15.06	0.039
P		4.0	3.9	4.0	3.5	4.1	3.7	4.1	3.6	4.4	4.1	3.9	3.6	4.2	3.5	4.0	3.7	4.2	4.1	3.7	3.9	4.55	5.93	0.005
K	g/kg	5.5	5.4	6.1	5.9	5.6	4.7	5.8	5.8	5.7	6.1	5.2	5.4	5.7	4.0	5.7	5.8	5.4	5.7	5.0	5.2	5.62	11.58	0.405
S		1.3	1.5	1.5	1.7	1.5	1.4	1.5	1.2	1.5	1.3	1.5	1.7	1.5	1.4	1.6	1.2	1.6	1.5	1.4	1.3	5.88	12.77	0.293
Mg		1.7	1.7	1.8	1.8	1.7	1.4	1.8	1.4	1.8	1.7	1.6	1.7	1.9	1.6	1.7	1.7	1.8	1.9	1.7	1.7	4.86	9.50	0.136
Na		16.6	18.9	26.4	17.7	26.8	35.0	33.1	26.4	35.0	27.1	24.2	38.8	26.7	21.4	13.8	13.9	24.6	26.5	30.1	21.4	25.62	31.29	0.755
Fe	mg/kg	40.3	42.3	42.3	42.5	40.8	32.0	42.9	30.5	44.4	41.8	39.9	37.6	40.9	30.0	46.3	43.3	39.7	40.7	41.1	42.8	5.10	14.18	0.083
Mn		59.6	53.9	53.5	53.5	60.3	38.0	44.1	38.6	59.7	53.3	47.9	49.4	46.0	42.0	45.9	42.1	51.4	45.7	47.3	52.5	12.28	13.63	0.119
Zn		30.5	27.2	33.3	35.7	30.8	20.0	31.9	25.2	34.9	23.9	30.3	32.3	24.1	30.0	33.8	28.1	28.9	24.0	29.9	29.1	9.80	16.51	0.078

[1]CV: coefficient of variation.

Table 8 *In vitro* dry matter, crude protein and starch digestibility (%) and viscosity (cP)[1] of triticale cultivars harvested in different years

Item	AT528		H20		H127		H128		H157		H249		H418		H426		JRCT74		Tahara		CV%[2]		P
	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	2008	2009	
Dry matter	76.95	74.20	75.28	75.38	76.30	75.44	77.50	76.79	74.01	76.47	76.47	77.34	77.63	76.23	77.36	74.56	76.86	74.41	75.38	75.48	1.52	1.40	0.154
Crude protein	57.12	55.85	57.54	52.17	49.38	52.22	53.11	49.98	51.81	39.73	42.15	60.72	48.73	42.70	37.87	43.86	42.67	48.92	40.79	50.46	14.44	12.65	0.609
Starch	83.83	87.80	84.11	83.97	87.18	85.41	85.09	87.51	85.41	84.59	84.61	83.59	84.75	87.43	87.81	86.81	86.54	84.01	85.84	85.24	1.54	1.89	0.861
Viscosity	1.04	1.03	1.09	1.01	1.08	1.04	1.03	1.03	1.02	1.02	1.04	1.09	1.08	1.05	1.02	1.06	1.03	1.04	1.01	1.03	2.75	2.17	0.734

[1] cP (in centipoises) = 1 mPa s (milliPascal second) = 1/100 dyne-second per square centimetre (dyn·s/cm²);
[2] CV: coefficient of variation.

variance between cultivars in each harvest year. For Na, the content ranged from 13.8 (H426; 2008) to 38.8 mg/kg (H249; 2009), whereas Fe content varied from 30.0 (H418; 2009) to 46.3 mg/kg (H426; 2008). The concentration of Mn was between 38.0 (H127; 2009) and 60.3 mg/kg (H127; 2008) and Zn content was from 20.0 (H127; 2008) to 35.7 mg/kg (H20; 2008). The concentrations of Na, Fe, Mn and Zn were not different between the two harvest years or between cultivars in each harvest year.

Table 8 shows the *in vitro* digestibility of DM, ranging from 74.0% (H157; 2008) to 77.6% (H418; 2008), digestibility of CP values was between 37.9% (H426; 2008) and 60.7% (H429; 2009) and digestibility of starch varied between 83.6% (AT528; 2009) to 87.8% (AT528; 2008). *In vitro* digestibility of DM, CP and starch was similar (*P* >0.05) between the two harvest years. The difference in digestibility between the cultivars was also low (CV <15%). In addition, the viscosity of the digesta ranged from 1.01 to 1.09 cP. Viscosity of the same cultivars harvested in 2008 was about the same (*P* >0.05) as that of samples harvested in 2009, with a CV less than 3%.
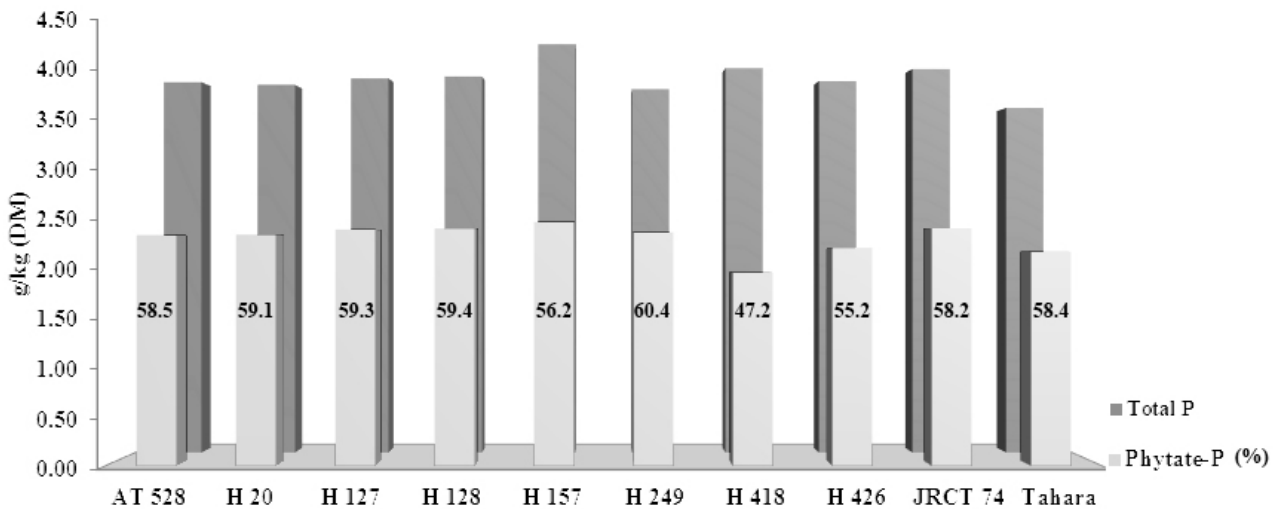
Figure 1 Proportion of phytate-P to total phosphorus (P) in triticale cultivars harvested in 2008.
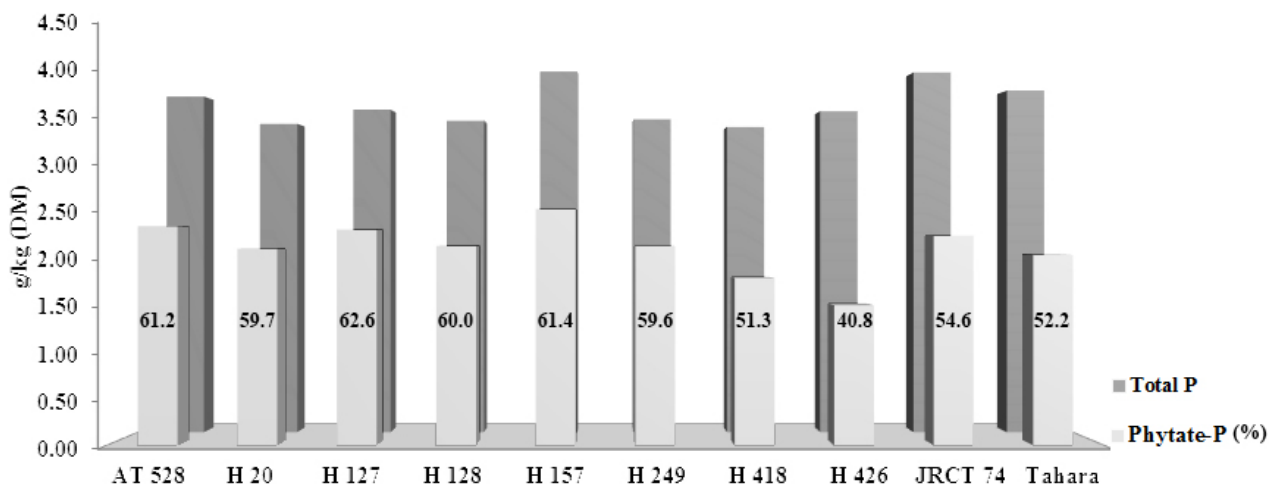
Figure 2 Proportion of phytate-P to total phosphorus (P) in triticale cultivars harvested in 2009.

The phytate-P content of the triticale cultivars harvested in 2008 and 2009 are presented in Figures 1 and 2, respectively. The proportion of phytate-P to total P in the samples harvested in 2008 ranged from 47.2% to 60.4%, whereas for the samples harvested in 2009 was between 40.8% and 62.6%. Similar to P

content, the phytate-P concentration was different (P = 0.08) between the two harvest years, although the variability of phytate-P concentration was relatively low in each year (6.67% and 13.88%, in year 2008 and 2009, respectively). Furthermore, the concentration of phytate-P varied between 1.96 (H418) and 2.48 g/kg (H157) in 2008 and 1.80 (H418) and 2.54 g/kg (H157) in 2009.

A sufficient number of representative granules was not measured to enable detailed statistical analysis, but there appeared to be little difference between the cultivars in starch granular structure. The shape of the starch granules was also similar to that of the parent grains, wheat and rye, which tend to show a bimodal distribution of granule sizes in the mature endosperm of the seed, corresponding with large A- and small B-type starch granules.

Discussion

The nutrient composition of grains is affected by many factors, including grain variety (genetic factors), growing location (agronomic conditions) and the season or environmental factors (O'Brien, 1999). However, in this study, nutrient composition was similar between the triticale cultivars in two consecutive harvesting years. Such similarities between grain batches have been reported by researchers working at different locations on different cultivars of triticale (Radcliffe *et al.*, 1983 Adeola *et al.*, 1986; Heger & Eggum, 1991; Salmon *et al.*, 2002; Brand *et al.*, 2003). Triticale appears to have genetic uniformity between the cultivars, although breeding programmes are still keen to improve their quality.

The DM content of triticale in the current study was less than values reported by Radcliffe *et al.* (1983), Salmon *et al.* (2002) and Brand *et al.* (2003), which was about 890 g/kg DM or more, but this may be the result of differences in varieties as well as the growth location of the triticale used in the studies. The triticale cultivars used in the current study were the spring varieties, popular in Australia (Cooper *et al.*, 2004) whereas the results reported by Heger & Eggum (1991) and Brand *et al.* (2003) were for winter varieties studied in Czechoslovakia and South Africa, respectively, as well as Adeola *et al.* (1986) and Salmon *et al.* (2002) in Canada. Other factors that can possibly affect the nutrient composition are weather, management, postharvest storage, pathogens and pests (Morris, 2004).

The protein content of triticale cultivars in this study was about 92 to 128 g/kg DM, lower than that reported by Brand *et al.* (2003) and Salmon *et al.* (2002), which was 132 to 159 g/kg DM, but relatively similar to Radcliffe *et al.* (1983) and Adeola *et al.* (1986). In comparison to wheat, McKenzie & Farrell (1980) and Flores *et al.* (1994) have reported that triticale has a higher protein value. The crude fat and ash contents of the triticale cultivars in the current study were relatively more variable than those in other studies, which were ranging from 13.9 to 27.4 g/kg DM and 16.7 to 20.0 g/kg DM, respectively. With regard to the GE content, there was only a minor variation between the cultivars as well as between the years of harvest. The values are also similar to those obtained in South Africa and Canada (Adeola *et al.*, 1986; Brand *et al.*, 2003), which were ranging from 18.1 to 18.5 MJ/kg DM.

In previous studies, starch contents of triticale varied from 630 to 730 g/kg DM (Leon *et al.*, 1996; Hughes & Cooper, 2002; Çiftci *et al.*, 2003). The contents found in the current study were lower than these values, the highest starch content being in cultivar H127, a trial name of triticale cultivar that was later commercialised as 'Bogong'. Although all the triticale samples from the two harvest years contained less total starch than was found in earlier studies, the lower content of resistant starch found in the triticale samples demonstrated that a large proportion of starch fraction may be digestible.

Moran (1982) and Tester *et al.* (2004) defined a 'normal' amylose content as being between 160 and 350 g/kg, and the starch is regarded as 'waxy' or 'amylo-' when the amylose content exceeds 350 g/kg. Based on these definitions, the composition of amylose and amylopectin of the cultivars of H249, AT528, H20, H127, H128 and H157 can be categorised as 'normal' while H418, JRCT74, H426, and Tahara cultivars, which contained less than 160 g/kg amylose are 'waxy' cultivars. Cultivar H418 is a trial name of a cultivar that is now commercialised as 'Canobolas'. In terms of amylose content, all the cultivars are potentially of high digestibility in view of their 'low' amylose contents. Oates (1997) and Carré (2004) postulated that starch with high amylose content (>40%) is often considered to be of lower digestibility, i.e. the starch becomes more resistant.

Triticale is more similar to wheat than rye in terms of NSP content. The major fraction of cereal NSP is arabinoxylans (O'Brien, 1999; Dervilly-Pinel *et al.*, 2001). The NSP level has a negative correlation with carbohydrate digestibility (Leeson & Summers, 2001). Austin *et al.* (1999) stated that water-insoluble NSP are indigestible by birds whereas soluble NSP are potentially digestible. However, Graham *et al.* (1993) reported that soluble NSP increase digesta viscosity, which can affect the growth and performance of birds. The new high-yielding triticale cultivars tested in the current study might have less problems of viscosity owing to their low percentage of water-soluble NSP. In addition, the NSP found in the triticale cultivars was mostly insoluble. Data obtained in this study suggest that the concentration of soluble NSP was five times less than that of insoluble NSP. Cultivar AT528 and Tahara had the highest soluble NSP of the cultivars,

whereas H128 and H20 contained the highest level of insoluble NSP. In comparison with NSP content of conventional ingredients used in poultry diets, such as maize and soybean meal, reported by Meng & Slominski (2005), four of triticale cultivars (H426, H418, H157 and H249) can be classified as low water-soluble NSP cultivars, whereas the other cultivars were slightly higher in NSP content than maize and soybean meal. Based on the total NSP found in the current study, the most common sugars were arabinose and xylose. Digestibility of triticale by birds therefore would likely be beneficial if they were supplemented with microbial enzymes targeting arabinans and xylans, which are the polymers of arabinose and xylose, respectively.

The AA profiles of ingredients are important in formulating poultry diets because supplemental AA are relatively expensive (D'Mello, 2003) and a balanced AA profile in the final diet will have a protein-sparing effect (Brandt *et al.*, 2000). The concentrations of all AA found in the current study were higher than those reported by Sikka *et al.* (1978), Huet *et al.* (1988) and Mosse *et al.* (1988). In addition, the AA concentrations found in these triticale cultivars were also higher than those reported by NRC (1994).

In most cereal grains, lysine is the 'first limiting' AA (Brown, 1989). These results showed that the lysine content of triticale cultivars (3.8 to 4.8 g/kg DM) was higher than for wheat or rye. This finding is in agreement with the results reported by Stallknecht *et al.* (1996). Moreover, Heger & Eggum (1991) and Mosse *et al.* (1988) reported that lysine levels did not decrease with decreasing protein content and have been reported to be proportionally higher when the protein content of grain was low. Furthermore, the content of lysine in some triticale cultivars was higher in the current study than reported by Flores *et al.* (1994) and Van Barneveld (2002). In the present study, the highest lysine concentration was in the H249 cultivar, while the lowest was in H128. Elliot (1995) concluded that, after lysine; methionine, threonine, tryptophan, isoleucine and arginine are the next limiting AA in most poultry diets. Tryptophan was not quantified in this study but the concentrations of other potentially limiting AA were high in cultivars H157, H127, H418 and H249.

With regard to the major elements, K content was the highest in the triticale cultivars investigated, followed by P, Mg, S, Ca and Na. The concentration of Mn was the highest among the trace elements, followed by Fe and Zn. The similarity in mineral composition between the cultivars as well as between triticale harvested in different years confirms the consistency of the cultivars tested. The difference in P between 2008 and 2009 may be due to external factors during the growing period, such as water supply, soil type and fertiliser. However, there was low variability in P content between the cultivars. In comparison with previous studies, the range of individual mineral contents corresponded with values reported by Lorenz *et al.* (1974) and Sehgal *et al.* (1983). Variations in concentration of mineral elements can be attributed to the effects of soil, water supply, time of sowing, fertilisers, as well as varietal differences. The values in this study indicate that triticale contains relatively higher amounts of the major elements K, P and Na than wheat. The Ca content, on the other hand, is relatively similar to wheat. In the same way, the important minor elements such as Zn, Cu, Fe and Mn were found to be present in higher amounts in triticale than in wheat as also reported by Lorenz *et al.* (1974).

The concentrations of phytate-P of triticale cultivars harvested in 2008 and 2009 were relatively similar. The levels of phytate-P were lower than those reported by Singh & Sedeh (1979) in a study of 14 cultivars of triticale, but they were in the range reported by Singh & Reddy (1977). In comparison, Kim *et al.* (2002) reported that the proportion of phytate-P to total P in wheat ranged from 55% to 79% in two harvest years. Extra attention should be paid to H157 cultivar when it is included in diets due to its high phytate-P content.

In the present study, *in vitro* digestibility of DM, starch and CP as well as the digesta viscosity were not different between the cultivars, or between the harvest years. These findings are positive for the rapid selection and use of the cultivars in diet formulation. However, the digestibility of starch is relatively lower than that in the study reported by Flores *et al.* (1994). The presence of anti-nutritive factors, particularly NSP, may be responsible for the low digestibility of starch and protein. The inclusion of exogenous enzymes such as phytase and/or carbohydrase could help to increase the digestibility of these nutrients. Viscosity values during digestion were relatively similar between the cultivars and this was mainly the result of the similarity in content and types of the NSP.

Conclusion

It may be concluded from the present study that the nutrient compositions of the 10 cultivars of triticale, harvested in two different years, were generally similar. Phosphorus content was the most variable between the two harvest years. The cultivars were superior, in terms of concentration of key nutrients such as protein and amino acids, to wheat and rye, the cereals from which triticale was bred.

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