Evaluation of Broiler Performance When Fed Roundup-Ready Wheat (Event MON 71800), Control, and Commercial Wheat Varieties[1]

C. A Kan*[,2] and G. F. Hartnell†

*Animal Sciences Group, Nutrition and Food, 8200 AB, Lelystad, The Netherlands; and †Monsanto Company, St. Louis, Missouri 63167

ABSTRACT We evaluated the nutritional value of broiler diets containing approximately 40% wheat grain from Roundup Ready wheat (MON 71800), its similar nontransgenic control (MON 71900), or reference commercial wheat varieties. The feeding trial lasted 40 d, and each treatment consisted of 10 replicates of 1-d-old Ross 308 broilers (5 pens of males and 5 pens of females). Each pen contained 12 birds, and at d 13 birds were randomly removed until 9 birds remained. Body weight and feed intake were measured on pen basis at 40 d. At d 41, four broilers per pen were slaughtered. The carcasses were dissected, and cut-up yields were determined. Dry matter, protein, and fat contents of breast meat were determined. The data were analyzed by an ANOVA procedure. The BW and feed conversion at d 40 averaged 2,450 g and 1.52, respectively. There were no significant treatment × sex interactions, except for evisceration yield with significant differences ($P < 0.05$) in yield between birds fed 2 commercial wheat varieties. Data for final BW, feed conversion, carcass yield, and breast meat were not statistically different ($P < 0.05$) between broilers fed MON 71800 or MON 71900 or the population of birds fed commercial wheat varieties, except a lower carcass yield at d 41 for birds fed the nontransgenic control wheat. Thus MON 71800 was nutritionally equivalent to nongenetically modified wheat varieties when fed to broilers.

(*Key words*: broiler, carcass yield, nutritional value, Roundup Ready wheat grain)

2004 Poultry Science 83:1325–1334

INTRODUCTION

Glyphosate is the active ingredient in the Roundup family of agricultural herbicides. Glyphosate-tolerant traits have been introduced into a number of crops, including wheat. The glyphosate-tolerant wheat evaluated in broilers was defined as Roundup Ready wheat containing event MON 71800 (or simply MON 71800). This event was generated by the insertion of a cassette containing the 5-enolpyruvylshikimate-3-phosphate synthase coding sequence (*cp4 epsps*) into the wheat genome of the Bobwhite cultivar. The *cp4 epsps* coding sequence, derived from the *Agrobacterium sp.* strain CP4, encodes for the production of the CP4 EPSPS protein, which confers tolerance to glyphosate.

Wheat is a source of amino acids and energy in broiler diets. Because broilers are very fast growing birds, any unintended changes in nutrient or antinutrient content will likely be reflected in their performance and carcass characteristics. This study is the first to evaluate the effect of feeding broilers with diets containing MON 71800 as compared with MON 71900 and 8 nontransgenic commercial wheat varieties. The goal of a study of substantial equivalence is to determine whether the transgenic product is substantially equivalent (in terms of chemical and nutritional composition and characteristics) to its conventional counterpart that has a history of safe use. Authorities then use this information in their decision-making process regarding the safety evaluation of crops derived from modern biotechnology. The results of this study should support nutritional equivalence of Roundup Ready wheat with its nontransgenic counterparts.

MATERIALS AND METHODS

Feedstuffs

The test substance was Roundup Ready wheat containing event MON 71800. The control substance was the parental nontransgenic Bobwhite cultivar line, designated as MON 71900, which has background genetics representative of the test event. In addition, nontransgenic commercial varieties of wheat were included in this study as reference controls. Wheat was included from 8 commercial reference varieties (Express, Cavalier, Hank, Westbred 926, McNeal, Ernest, Fortuna, and Westbred 936).

Received for publication December 19, 2003.
Accepted for publication February 18, 2004.
[1]Roundup Ready and Roundup are registered trademarks of Monsanto Technology LLC.
[2]To whom correspondence should be addressed: kees.kan@wur.nl; kan@dronten.net.

TABLE 1. Analyses of nutrients and minerals in the 10 wheat varieties (as-is basis)

Variety	MON 71800[1]	MON 71900[2]	Express	Cavalier	Hank	Westbred 926	McNeal	Ernest	Fortuna	Westbred 936
Dry matter (g/kg)	913	909	910	905	909	903	907	904	904	892
Ash (g/kg)	22	20	23	21	25	27	26	22	19	17
Crude protein (g/kg)	168	153	171	168	179	174	170	195	166	200
Crude fat (g/kg)	19	21	24	18	19	21	16	20	22	20
NDF[3] (g/kg)	95	95	105	92	97	112	107	119	109	114
ADF (g/kg)	39	41	51	23	36	45	44	45	44	38
Lignin (g/kg)	8	13	3	1	7	3	12	12	12	8
NSC (g/kg)	612	625	591	608	593	572	591	551	590	544
Ca (g/kg)	1.0	0.8	0.7	0.6	0.8	0.8	0.6	0.7	0.4	0.5
P (g/kg)	5.0	5.2	5.8	5.2	5.7	5.7	4.6	5.7	4.9	4.5
Mg (g/kg)	1.5	1.5	1.7	1.5	1.7	1.7	1.5	1.8	1.7	1.5
K (g/kg)	4.9	4.9	5.0	4.8	6.0	5.2	4.2	5.2	4.7	3.9
Na (g/kg)	0.04	0.06	0.04	0.04	0.06	0.04	0	0.01	0	0
Sulfur (g/kg)	1.5	1.4	1.5	1.5	1.6	1.5	1.6	1.8	1.7	1.8
Fe (mg/kg)	64	48	40	102	54	50	124	59	128	41
Zn (mg/kg)	36	34	32	23	37	31	30	68	35	21
Cu (mg/kg)	4	3	2	2	5	3	3	4	3	1
Mn (mg/kg)	32	36	41	20	36	37	29	35	40	27
Mo (mg/kg)	0.5	0.4	0.4	0.6	0.5	0.5	0.4	0.5	0.6	0.2

[1]MON 71800 is Roundup Ready wheat.
[2]MON 71900 is the parental nontransgenic cultivar.
[3]NDF = neutral detergent fiber; ADF = acid detergent fiber; NSC = nonstructural carbohydrates.

All lines were grown in the US in the 2000 growing season. All batches of wheat were ground using the same screen size (1/8-inch screen) before shipping to the Netherlands. The wheat was stored at 4 to 8°C before and after grinding, shipped at room temperature, and stored 4 to 8°C at ID-Lelystad in the Netherlands. Accountability for test, control, and reference substance was maintained throughout the study. Samples were taken to measure glyphosate residue levels in the Roundup Ready wheat and its parental nontransgenic control cultivar.[3]

Representative subsamples of each test, control, and reference wheat material were taken and analyzed for the following components: moisture, crude protein, crude fat, ash, crude fiber, carbohydrates, acid detergent fiber, neutral detergent fiber, Ca, P, K, Na, Mg, S, Zn, Fe, Mn, and Cu;[4] amino acids;[5] mycotoxins;[6] and pesticides.[5] Soybean and tapioca to be used in diet preparation were sampled for analyses of nutrient content[4] and screened for mycotoxins[6] and pesticides.[5] Analyses of ingredients and feeds at ID-Lelystad included moisture, crude fiber, crude protein, crude fat, amino acid composition (except tapioca), starch, ash, Ca, P, Mg, K, Na, S, Mn, Fe, Cu, and Zn.

Experimental Feeds

Sources of dietary protein in the experimental diet were wheat and solvent-extracted dehulled soybean meal and supplemented with lysine, methionine, and threonine to meet nutritional requirements. All diets were formulated to contain approximately equal amounts of the 3 first limiting dietary essential amino acids (methionine, cystine, and lysine), Ca, absorbable P (Dutch feed tables; Centraal Veevoederbureau, 1998), and sodium based on analytical data from the feedstuffs. All diets also contained similar levels of AME content in starter (11.9 MJ/kg) and finisher (12.4 MJ/kg). All diets conformed to industry standards and met or slightly exceeded nutritional recommendations (Centraal Veevoederbureau, 1998). Compositions of the different diets varied only slightly among the wheat varieties. The results of chemical analyses of the 10 wheat varieties are listed in Table 1. The dietary compositions and calculated nutrient contents for all starter and finisher diets are listed in Tables 2 and 3. The amounts of added micronutrients are specified in Table 4. The starter diets contained between 393 and 402 g/kg of the different wheat varieties, and the finisher diets contained between 395 and 399 g/kg wheat. All diets contained a coccidiostat (Lerbek, a mixture of meticlorpindol and methylbenzoquate[7]) at 110 mg/kg of feed. The diets did not contain any growth promotants. Analyzed contents of most nutrients are listed in Tables 5 and 6.

Treatment diets were mixed and pelleted through a 3-mm die with live steam addition at Arkervaart feed mill in Leusden (The Netherlands). Tapioca and soybean meal were ground through a 2.5- to 3-mm screen prior to mixing.

After being pelleted, feed was continuously subsampled when leaving the cooler prior to bagging the feed. This large subsample was mixed, and duplicate samples of 500 g each were taken. One sample was subdivided for nutrient analysis at ID-Lelystad, and the second sample was stored between –13 and –26°C.

Dry matter and ash contents were determined according to AOAC (1984). Nitrogen was assayed by the

[3]Monsanto Company, St. Louis, MO.
[4]Dairy One, Ithaca, NY.
[5]Covance Laboratories, Madison, WI.
[6]Romer Laboratories, Union, MO.
[7]Merial www.merial.com.

TABLE 2. Ingredients and calculated nutrient composition of the experimental starter diets (g/kg of diet; as-is basis)

Item	Express	MON 71800[1]	MON 71900[2]	Cavalier	Hank	Westbred 926	McNeal	Ernest	Fortuna	Westbred 936
Wheat variety	A	B	C	D	E	F	G	H	I	J
Soybean meal	321	321	321	319	320	321	318	319	318	317
Tapioca meal	160	160	160	160	160	160	160	170	160	170
Animal fat	80	80	80	80	80	80	80	80	80	80
Limestone	14.7	13.9	14.3	14.5	14.5	14.5	14.2	14.5	14.5	14.1
Monocalcium phosphate	9.8	10.7	10.4	10.4	9.9	9.9	11.1	10.0	10.8	11.3
NaCl	3.7	3.7	3.6	3.7	3.6	3.7	3.7	3.7	3.7	3.7
L-Lysine HCl	1.8	1.8	1.8	1.8	1.6	1.7	1.9	1.6	1.9	1.8
DL-Methionine	2.7	2.7	2.8	2.7	2.5	2.6	2.6	2.3	2.5	2.5
L-Threonine	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5
Vitamins and minerals	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
Wheat										
Wheat A/F	400	—	—	—	—	400	—	—	—	—
Wheat B/G	—	400	—	—	—	—	402	—	—	—
Wheat C/H	—	—	400	—	—	—	—	392	—	—
Wheat D/I	—	—	—	401	—	—	—	—	402	—
Wheat E/J	—	—	—	—	401	—	—	—	—	393
Total	1,000	1,000	1,000	1,000	1,000	1,000	1,000	1,000	1,000	1,000
Ash	65	65	64	65	66	67	67	65	64	64
Crude protein	219	217	214	219	225	221	219	227	218	229
Crude fat	96	94	94	93	94	94	92	94	95	94
Crude fiber	29	29	29	29	29	29	29	29	29	29
AME_n broilers (MJ/kg)	11.90	11.90	11.90	11.90	11.90	11.90	11.90	11.90	11.90	11.90
Ca	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0
P	6.9	6.8	6.8	6.8	6.9	6.9	6.7	6.9	6.8	6.7
Na	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5
Cl	2.9	2.9	2.9	2.9	2.8	2.9	2.9	2.8	2.9	2.9
Fecal digestible amino acids										
Lysine	10.6	10.6	10.6	10.6	10.6	10.6	10.6	10.6	10.6	10.6
Methionine	5.2	5.3	5.3	5.2	5.2	5.2	5.2	5.0	5.2	5.2
Methionine + cystine	8.0	8.0	8.0	8.0	8.0	8.0	8.0	8.0	8.0	8.0

[1]MON 71800 is Roundup Ready wheat.

[2]MON 71900 is the parental nontransgenic cultivar.

Dumas method (AOAC, 1984), and protein content was calculated as 6.25 × N. Crude fat was assayed by the Berntrop method (EU directive 84/4/EEG, methods A and B, 1984).[8] Fat analysis in breast meat was assayed by the petroleum-ether extraction method 40/60 (EU directive 84/4/EG, method A, 1984).[8] Crude fiber was assayed according to ISO/DP 6865.[9] The amino acids (except methionine, cystine, and tryptophan) were assayed by ion-exchange column chromatography after hydrolysis for 23 h in HCl (6 mol/L). Cystine and methionine were determined as cysteic acid and methionine sulfone after oxidation with performic acid before hydrolysis (Schram et al., 1954). Tryptophan was determined according to Sato et al. (1984). Total P, Ca, Mg, Cu, Zn, Fe, Na, Cl, and K were determined by atomic emission spectrometry according to ISO 11885 (ISO, 1996). Starch was assayed by the α-amylo glucosidase procedure with previous extraction as described by NEN 3574 (NEN, 1974).

Experimental Details

The study protocol was approved by the Animal Ethics Committee of ID-Lelystad, as requested by the Dutch Law on Animal Experiments. The study was carried out in Spring 2001 adhering to the approved protocol. The study included 1,200 normal, healthy 1-d-old Ross 308 chicks. Feather sexing was performed to distinguish between males and females. All birds were vaccinated for infectious bronchitis and Newcastle disease on d 1, Gumboro disease on d 20, and Newcastle disease on d 21.

Birds were housed in 100 floor pens on concrete (dimensions 1 m × 0.75 m with 0.7 to 0.8 m wire walls) in an environmentally controlled facility. All birds were placed in clean pens containing approximately 5 to 10 cm of wood shavings as bedding. Additional bedding was added to pens if needed. Lighting was via incandescent bulbs, and after 48 h of full lighting a program of 1L:3D was used. Environmental temperature (controlled by the process computer) was set at 33°C at d 1 and lowered stepwise to 20°C in wk 6.

Environmental conditions for the birds (i.e., floor space, temperature, lighting, bird density, feeder, and water space) were similar for all experimental groups. Assignment of the 10 treatments to the 100 pens was conducted to provide a randomized complete block design with five blocks (5 pens of male birds and 5 pens of female birds per treatment group with each treatment represented in each block).

The trial started with 12 birds (males or females) per pen. On d 13, all birds within a pen were counted. If

[8]www.europa.eu.int.

[9]www.iso.ch.

TABLE 3. Ingredient and calculated nutrient composition of the experimental finisher diets (g/kg of diet; as-is basis)

Item	Express	MON 71800[1]	MON 71900[2]	Cavalier	Hank	Westbred 926	McNeal	Ernest	Fortuna	Westbred 936
Wheat variety	A	B	C	D	E	F	G	H	I	J
Soybean meal	297	297	297	297	297	297	295	296	295	294
Tapioca meal	180	180	180	180	180	180	180	180	180	185
Animal fat	95	95	95	95	95	95	95	95	95	95
Limestone	11.0	10.3	10.6	10.8	10.9	10.9	10.5	11.0	10.9	10.5
Monocalcium phosphate	4.6	5.5	5.2	5.2	4.7	4.7	5.9	4.7	5.6	6.1
NaCl	3.1	3.1	3.1	3.1	3.1	3.1	3.2	3.2	3.2	3.2
L-Lysine HCl	2.1	2.1	2.2	2.2	1.9	2.0	2.2	1.9	2.2	2.1
DL-Methionine	2.7	2.7	2.8	2.7	2.5	2.6	2.6	2.3	2.5	2.5
L-Threonine	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5
Vitamins and minerals	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
Wheat										
Wheat A/F	398	—	—	—	—	398	—	—	—	—
Wheat B/G	—	398	—	—	—	—	399	—	—	—
Wheat C/H	—	—	398	—	—	—	—	399	—	—
Wheat D/I	—	—	—	398	—	—	—	—	399	—
Wheat E/J	—	—	—	—	398	—	—	—	—	395
Total	1,000	1,000	1,000	1,000	1,000	1,000	1,000	1,000	1,000	1,000
Ash	57	57	56	56	57	58	59	57	56	55
Crude protein	209	207	204	209	215	211	209	219	208	220
Crude fat	110	108	109	108	108	109	107	108	109	108
Crude fiber	29	29	29	29	29	29	29	29	29	29
AME_n broilers (MJ/kg)	12.40	12.40	12.40	12.40	12.40	12.40	12.40	12.40	12.40	12.40
Ca	6.7	6.7	6.7	6.7	6.7	6.7	6.7	6.7	6.7	6.7
P	5.6	5.5	5.5	5.5	5.6	5.6	5.4	5.6	5.4	5.4
Na	1.3	1.3	1.3	1.3	1.3	1.3	1.3	1.3	1.3	1.3
Cl	2.6	2.6	2.6	2.6	2.6	2.6	2.7	2.6	2.7	2.6
Fecal digestible amino acids										
Lysine	10.3	10.3	10.3	10.3	10.3	10.3	10.3	10.3	10.3	10.3
Methionine	5.1	5.1	5.2	5.1	5.0	5.1	5.1	4.8	5.0	5.0
Methionine + cystine	7.7	7.7	7.7	7.7	7.7	7.7	7.7	7.7	7.7	7.7

[1]MON 71800 is Roundup Ready wheat.

[2]MON 71900 is the parental nontransgenic cultivar.

more than 9 birds were present, any unthrifty (apparently disabled or low weight) birds were removed. Next, other birds were randomly removed based on the highest wing tag number until the required number was reached; this procedure allowed sufficient space for each bird at d 40. Removed birds were killed by suffocation with CO_2.

TABLE 4. Addition of vitamins and minerals per kilogram of diet

Item	Addition (/kg of diet)	
Vitamin A	10,000	IU
Vitamin D_3	2,000	IU
Vitamin E	30	IU
Vitamin K_3	1.5	mg
Vitamin B_1	2.0	mg
Vitamin B_2	7.5	mg
Pantothenic acid	10	mg
Niacin	35	mg
Vitamin B_{12}	20	μg
Folic acid	1	mg
Biotin	150	μg
Vitamin B_6	3.5	mg
Choline	460	mg
Fe	50	mg
Cu	10	mg
Zn	40	mg
Mn	60	mg
I	0.8	mg
Co	0.3	mg
Se	0.1	mg
Meticlorpindol	100	mg
Methylbenzoquate	8.4	mg
Anti-oxidant E310,320,321	50	mg

Feed was provided ad libitum throughout the study via one feeder per pen. A chick feeder tray was placed in each pen for the first 2 d. All birds were placed on their respective diets from d 0 onward, and diets were fed continuously for 40 d at which time the feeding part of the study was terminated. All feed added and removed from pens was weighed. The feeding periods for the starter and finisher diets were 0 to 13 d and 13 to 40 d, respectively, for all pens.

Water was provided ad libitum throughout the study via 2 automatic drinkers (drinking cups) per pen connected to a central watering system. For the first 2 d, a water tray was provided in each pen. The test facility, pens, and birds were observed twice daily during weekdays and once daily during weekends for general flock condition, lighting, water, feed, ventilation, and any unanticipated events. Any bird that was removed, found dead, or killed was weighed and recorded on the pen mortality record. All mortalities were necropsied to determine the probable cause of death. Probable initial cause of death and necropsy findings were recorded.

Birds were weighed by pen during the study on d 0, 13, 35, and 40 (end of the feeding study). Pens were weighed in successive order within a block. Feed bins were weighed at the same time.

TABLE 5. Dry matter, ash, crude protein, crude fiber, crude fat, starch, N-free extractives (g/kg diet; as-is basis) and minerals (mg/kg) in the experimental diets

Diet	Treatment	Wheat variety	Dry matter	Ash	Crude protein	Crude fiber	Crude fat	Starch	N-free extract[1]	Ca	Mg	P	Na	K	Cl	Cu	Zn	Fe
			Experimental starter diets from d 1 through 13															
01	01	Express	898	63	226	32	100	286	477	9.3	2.2	6.4	1.7	10.2	3.2	15	73	600
02	02	MON 71800[2]	895	62	221	30	97	283	484	9.4	2.1	6.6	1.7	10.0	3.0	17	89	614
03	03	MON 71900[3]	896	62	221	32	98	290	484	9.3	2.2	6.5	1.6	10.3	3.1	18	79	617
04	04	Cavalier	896	63	229	32	96	290	476	8.9	2.1	6.3	1.5	10.2	3.0	15	71	613
05	05	Hank	898	63	227	31	98	280	478	9.3	2.2	6.4	1.6	10.4	3.0	17	79	619
06	06	Westbred 926	894	64	228	31	100	271	470	9.4	2.2	6.5	1.6	10.3	3.1	16	73	597
07	07	McNeal	889	61	224	31	99	269	476	9.5	2.2	6.4	1.7	9.8	3.1	17	77	606
08	08	Ernest	894	63	235	31	100	252	466	8.9	2.2	6.3	1.5	10.3	3.1	16	83	614
09	09	Fortuna	891	63	228	33	101	287	467	9.2	2.1	6.2	1.6	9.7	3.2	17	76	649
10	10	Westbred 936	888	61	234	32	98	270	464	9.1	2.1	6.2	1.5	9.7	2.9	16	69	615
			Experimental finisher diets from d 13 through 40															
11	01	Express	892	55	211	31	110	286	485	6.6	2.0	4.9	1.4	9.5	2.8	14	69	613
12	02	MON 71800[2]	896	54	207	30	110	320	494	6.3	2.0	5.0	1.2	9.3	2.6	14	74	754
13	03	MON 71900[3]	898	54	208	30	109	311	497	6.4	2.0	4.8	1.3	9.3	2.8	15	73	685
14	04	Cavalier	895	54	213	29	109	283	490	6.6	2.0	5.0	1.3	9.6	2.7	14	68	654
15	05	Hank	897	56	213	30	113	270	486	6.6	2.0	4.9	1.3	9.9	2.7	19	73	649
16	06	Westbred 926	892	55	212	35	112	271	478	6.7	2.1	5.1	1.3	9.7	2.7	18	66	624
17	07	McNeal	888	54	209	29	110	288	485	6.5	2.0	4.8	1.4	9.1	2.6	15	70	598
18	08	Ernest	891	55	219	30	113	265	474	6.6	2.0	4.8	1.3	9.3	2.8	18	82	638
19	09	Fortuna	886	54	211	30	112	283	479	6.7	2.0	5.0	1.3	8.8	2.9	17	72	641
20	10	Westbred 936	886	54	219	31	112	292	471	6.7	1.9	5.0	1.3	8.8	2.6	16	66	682

[1]Nitrogen-free extractives = organic matter – (crude protein + crude fat + crude fiber).
[2]MON 71800 is Roundup Ready wheat.
[3]MON 71900 is the parental nontransgenic cultivar.

Carcass Traits

Birds remained on diet until the end of d 40. Feed was withdrawn at 2300 h onward, and about 12 to 16 h later, 4 birds with the lowest wing tag numbers in each pen were slaughtered with standard industrial machinery under experimental conditions. The birds were weighed and then hung by their legs on the slaughter line and stunned by an electric current (>50 mA per bird). The carotid artery and vein were severed, and after bleeding the birds were submerged in a scalding tank (approximately 51°C) for 4 to 5 min. They were then mechanically plucked, and the feet and heads were manually removed. The intestinal package was removed mechanically, and the remaining was carcass weighed. The carcasses were then chilled (approximately 2°C), and on one of next 3 d they were dissected to determine yields of breast meat, thighs, drumsticks, wings, rest of carcass, and carcass yield according to Uijttenboogaart and Gerrits (1982). Remaining birds were killed by T61[10] (Embutramide, 200 mg/mL; mebezoniumiodide, 50 mg/mL; and tetracaine hydrochloride, 5 mg/mL) injection, and all carcasses were incinerated. The right breast muscle of each slaughtered bird was collected for analysis of fat, protein, and dry matter contents on a composite sample per pen.

Performance data were summarized by average weight per bird on d 0 and 40 and by total feed intake. Adjusted feed conversion was calculated by using total feed consumption minus the assumed feed consumption of the dead or removed birds in a pen divided by the total growth of the surviving birds at the end of the study.

Data Analysis

For the 10 experimental treatments, male and female broilers were distributed randomly per block over the experimental units. The observations concerning the performance of the broilers, carcass yield of the broilers, and chemical analysis in breast meat of the broilers were statistically analyzed by ANOVA (GENSTAT, 1998) according to the following models.

In the first model, all treatments were compared in a pairwise fashion with each other.

$$\text{Model 1: } Y = \mu + \text{block}_i + T_j + S_k + T \times S_{jk} + \text{error}_{ijk}$$

where Y = response parameter (live performance, carcass yield, and chemical analysis in breast meat), μ = overall mean, block = effect of block (i = 1…5), T = treatment (experimental diet) effect (j = 1…10), S = sex effect (k = 1,2), and error = error term, which is assumed to be independent and normally distributed with mean equal to zero and variance to σ^2.

In the second model, the test group and nontransgenic control were each compared with the population of commercial reference wheat-fed groups.

$$\text{Model 2: } Y = \mu + \text{block}_i + V_l + T(V) + S_k + T(V) \times S + S \times V_{kl} + \text{error}_{ijklm}$$

where Y = response parameter (live performance, carcass yield, and chemical analysis in breast meat), μ = overall

[10]Hoechst Holland NV, Amsterdam, The Netherlands.

TABLE 6. Analyzed content of amino acids (g/kg of diet; as-is basis)

	Experimental starter diets from d 1 through 13										Experimental finisher diets from d 13 through 40									
	01	02	03	04	05	06	07	08	09	10	11	12	13	14	15	16	17	18	19	20
Treatment	01	02	03	04	05	06	07	08	09	10	01	02	03	04	05	06	07	08	09	10
Wheat	A[1]	B[2]	C[3]	D[4]	E[5]	F[6]	G[7]	H[8]	I[9]	J[10]	A[1]	B[2]	C[3]	D[4]	E[5]	F[6]	G[7]	H[8]	I[9]	J[10]
Amino acid																				
Lysine	12.2	12.6	12.0	11.7	12.4	11.6	12.8	12.0	12.0	12.1	10.5	11.7	11.6	11.9	11.5	11.6	11.5	11.5	11.7	11.6
Methionine	5.4	5.2	5.4	5.3	5.2	5.5	5.3	5.2	5.2	5.4	5.1	4.9	5.1	5.0	4.8	5.0	5.0	4.9	5.0	4.9
Cystine	3.9	3.9	3.7	3.6	3.7	3.8	3.9	3.7	3.8	3.8	3.4	3.5	3.5	3.3	3.3	3.4	3.3	3.6	3.5	3.6
Threonine	8.9	9.6	8.6	9.0	9.6	8.8	9.3	9.1	8.8	9.2	8.4	8.4	8.2	8.8	8.7	8.7	8.9	8.9	8.5	8.7
Tryptophan	2.7	2.6	2.4	2.7	2.7	2.7	2.7	2.9	2.8	2.8	2.5	2.5	2.6	2.5	2.5	2.6	2.5	2.6	2.6	2.6
Isoleucine	9.2	9.6	9.0	9.0	9.5	9.2	9.7	9.6	9.4	9.5	8.8	8.5	8.7	8.5	8.5	8.5	8.6	8.6	8.4	8.6
Arginine	14.8	15.6	14.8	14.8	14.4	15.2	15.6	14.9	15.5	14.1	14.1	13.6	14.4	13.0	12.8	13.4	12.7	12.9	12.9	12.1
Phenylalanine	10.6	10.8	9.9	10.4	11.1	11.0	11.4	11.0	11.2	10.8	10.2	9.6	9.8	10.2	9.9	10.0	9.9	10.1	9.8	10.1
Histidine	6.2	7.2	6.0	6.2	7.1	6.3	6.3	6.7	6.1	6.5	5.9	5.4	5.9	5.6	6.3	5.8	6.2	6.0	5.9	5.9
Leucine	16.1	16.3	15.4	15.7	16.4	16.0	16.8	16.7	16.2	16.6	15.2	14.7	15.0	14.8	14.8	15.0	15.0	15.1	14.8	15.1
Tyrosine	8.0	8.6	7.7	7.8	8.4	8.1	8.4	8.2	8.3	8.4	7.7	7.5	7.7	7.0	7.7	7.6	7.7	7.6	7.6	7.8
Valine	10.1	10.7	9.1	9.2	10.6	10.1	10.5	10.4	10.3	10.4	9.0	9.4	8.9	9.8	9.5	9.5	9.6	9.5	9.4	9.5
Alanine	9.3	10.5	8.8	9.0	10.2	9.3	9.5	9.6	9.1	9.5	8.4	8.5	8.5	8.3	8.5	8.3	8.7	8.7	8.3	8.3
Aspartic acid	20.3	21.7	20.0	20.2	21.6	20.2	21.1	20.4	20.2	20.4	19.1	18.7	18.9	18.6	19.1	19.0	19.2	19.1	18.7	18.9
Glutamic acid	47.2	46.6	44.0	46.4	46.7	46.9	48.0	48.7	46.8	50.2	45.6	43.3	43.0	48.5	44.8	45.8	44.6	46.4	44.4	47.6
Glycine	9.4	11.4	9.0	9.5	11.0	9.5	9.8	10.0	9.3	10.0	8.7	8.6	8.7	9.0	9.4	9.0	9.6	9.2	8.8	9.1
Proline	15.0	16.0	15.2	16.1	15.4	14.8	16.2	16.5	15.4	17.7	14.4	13.8	15.1	14.6	12.9	13.6	13.7	14.5	13.9	14.1
Serine	11.3	14.9	10.6	11.6	14.5	11.2	11.5	12.3	11.1	12.2	10.5	9.9	10.5	10.6	12.0	10.8	12.6	11.7	11.0	11.2
Sum of amino acids	220	234	212	218	231	220	229	228	222	230	209	202	206	210	207	208	209	211	205	210

A[1] = Express, B[2] = MON 71800, C[3] = MON 71900, D[4] = Cavalier, E[5] = Hank, F[6] = Westbred 926, G[7] = McNeal, H[8] = Ernest, I[9] = Fortuna; J[10] = Westbred 936.

mean, block = effect of block (i = 1…5), V = variety type (transgenic, nontransgenic line, and reference wheat varieties) effect (l = 1…3), T (V) = random effects of wheat varieties within type (treatments 1 and 4 to 10), S = sex effect (k = 1,2), T(V) × S = interaction effect of treatment (within variety) and sex, S × V = interaction effect of sex and variety type, error = error term, which is assumed to be independent and normally distributed with mean equal to zero and variance to σ^2.

RESULTS AND DISCUSSION

General Observations

Analyses of the feedstuffs did not reveal the presence of mycotoxins or significant amounts of pesticides. Levels of mycotoxins were nondetectable. All values for the pesticide screen were below the assay limits of detection [organophosphates (0.050 mg/kg), organochlorines (0.200 mg/kg), organonitrogens (0.500 mg/kg) and N-methylcarbamates (0.100 mg/kg)] except for organophosphates in Ernest (0.829 mg/kg) and Fortuna (0.231 mg/kg). The residue levels of glyphosate were very low (<0.10 mg/kg) in MON 71800 and MON 71900. The amino acid analysis data on the test, control, and reference wheat grain samples were used to calculate compositions of the different diets to supply equivalent amounts of nutrients to the birds. The feed analysis data of the different diets (Tables 5 and 6) confirmed the achievement of the intended nutrient levels in the diet.

Bird Performance and Carcass Characteristics

Diet Comparisons. The birds grew well and showed a low feed-to-gain ratio. Data for BW at d 0 and 40, feed intake, and feed-to-gain ratios are summarized in Table 7. Mortality was very low (1.3%) with no apparent relationship between treatment group and mortality. Mortality from d 0 to 40 totaled 15 birds, which randomly occurred over all treatments (MON 71800: one bird; MON 71900: zero birds; Express: 2 birds; Cavalier: 4 birds; Hank: 2 birds; Westbred 926: one bird; McNeal: 2 birds; Ernest: one bird; Fortuna: one bird; and Westbred 936: one bird).

Because there were no treatment × sex interactions except for evisceration yield, data for males and females were combined. The BW at 40 d of age, feed intake, and feed conversion of the broilers did not differ significantly among treatments (Table 7). Initial BW data of the 1-d-old broilers were different ($P < 0.05$). This effect was not expected and is attributed to chance with no adverse impact on the outcome of this study. No differences ($P > 0.05$) were noted in carcass yield parameters at d 41 weight, slaughter yield, and breast and drum meat yields across treatments. The carcass yield parameters of evisceration yield, thigh yield, wing yield, and remaining carcass yield showed differences among treatments ($P < 0.05$). Birds fed MON 71800, MON 71900, and 6 of the 8 reference diets had similar evisceration yield; however, those fed diets containing the commercial reference varieties McNeal and Ernest had a significantly higher evisceration yield than birds fed MON 71800, MON 71900, and Westbred 926. Birds fed diets containing the commercial reference variety Ernest also had higher thigh yields compared with birds fed MON 71800, MON 71900, and multiple commercial references (Table 7). The wing yield of birds fed diets containing MON 71800 was similar ($P > 0.05$) to those fed MON 71900 and all reference diets. The wing yield of birds fed the MON 71900 was higher ($P < 0.05$) than that of birds fed diets formulated from commercial reference varieties Westbred 926, Ernest, and Westbred 936. Broilers fed diets with MON 71800 and MON 71900 did not differ ($P < 0.05$) from the other 8 reference treatments in yield of remaining carcass. No effect of treatments could be detected on the chemical analysis in breast meat (Table 7). The treatment × sex interaction was not significant, except for evisceration yield of birds. In birds fed the commercial reference varieties Hank and McNeal, the differences between sexes in evisceration yield were 0.78 and 1.44 (data not shown), which are greater than the least significant difference of 0.57 across all treatments (Table 7).

Test Vs. Population of Nontransgenic Lines. Comparison of broiler performance for birds fed a diet containing MON 71800 vs. birds fed diets containing MON 71900 and the population formed by the diets from the other nontransgenic commercial reference wheat varieties showed no significant differences, with one exception (Table 8). Although pairwise comparisons revealed no differences across treatments, the birds fed the MON 71800 and reference population of commercial varieties had a higher body weight at d 41 than birds fed the diet with MON 71900. The interaction effect of variety × sex was only significant for thighs and drumstick yield (data not shown). Male broilers fed commercial reference wheat varieties had significantly higher thigh and drumstick yields than females, whereas the thigh and drumstick yields of birds fed diets containing MON 71800 showed no significant differences between sexes. As expected, BW gain and feed intake of male broilers were significantly higher and feed conversion was significantly lower than that of females (Table 9). Slaughter yield, breast meat yield, and remaining carcass yield of female broilers were significantly higher than male broilers, whereas evisceration yield, thigh, and drumstick yields were significantly lower. The results of the wings yield and chemical protein and fat analyses in breast meat were similar for male and female broilers, whereas dry-matter analyses of breast meat were significantly higher in female broilers (Table 9).

The yield data for breast meat, thighs, and drumsticks are similar to those reported by Veerkamp and Rincker (Veerkamp and Rincker, 1990), who used a similar method.

Overall, there were no biologically relevant differences among the performance, carcass yield, and breast meat analytical parameters for broilers fed diets containing MON 71800 as compared with MON 71900 and multiple commercial reference varieties. Therefore, the nutritional

TABLE 7. Broiler performance, carcass yield, and chemical analysis in breast meat (mean values of males and females). Comparison of Roundup Ready wheat (MON 71800) with its parental nontransgenic control line (MON 71900) and 8 commercial reference wheat varieties[1]

	Treatment										Treatment effect (T)	Sex effect (S)	Interaction effect T × S			
	2	3	1	4	5	6	7	8	9	10						
Wheat	B	C	A	D	E	F	G	H	I	J	SSD[4]	LSD[5]	SSD[4]	LSD[5]	SSD[4]	LSD[5]
Monsanto ID	MON 71800[2]	MON 71900[3]	Express	Cavalier	Hank	Westbred 926	McNeal	Ernest	Fortuna	Westbred 936						
Performance[6]																
Weight (g) d 0	43.6^{a}	42.2^{c}	42.3^{c}	42.4^{c}	43.5^{ab}	43.1^{abc}	42.7^{abc}	42.6^{bc}	42.4^{c}	42.9^{abc}	*	0.89	NS	0.40	NS	1.26
Weight (g) d 40	2,464	2,418	2,446	2,480	2,464	2,477	2,446	2,460	2,460	2,498	NS	78.0	***	34.9	NS	110
Feed intake (g) 0–40 d	3,675	3,608	3,633	3,685	3,656	3,679	3,674	3,636	3,667	3,758	NS	119	***	53.4	NS	169
Feed conversion (g/g)	1.52	1.52	1.52	1.52	1.52	1.52	1.53	1.51	1.52	1.54	NS	0.020	***	0.009	NS	0.028
Carcass yield[7]																
Weight (g) d 41	2,430	2,307	2,405	2,391	2,415	2,361	2,426	2,420	2,408	2,441	NS	94.6	***	42.3	NS	134
Slaughter yield (%)	86.8	86.7	86.9	86.9	86.6	87.1	86.9	87.0	86.8	86.4	NS	0.50	***	0.22	NS	0.71
Evisceration yield (%)	87.5^{b}	87.6^{b}	87.9^{ab}	87.8^{ab}	87.7^{ab}	87.5^{b}	88.1^{a}	88.1^{a}	87.8^{ab}	87.9^{ab}	*	0.40	***	0.18	*	0.57
Breast meat yield (%)	19.6	19.4	19.7	19.6	19.8	20.1	19.8	19.8	19.6	20.0	NS	0.58	***	0.26	NS	0.82
Thighs yield (%)	14.3^{bc}	14.3^{bc}	14.1^{c}	14.3^{bc}	14.3^{bc}	14.4^{ab}	14.3^{bc}	14.6^{a}	14.4^{ab}	14.3^{bc}	*	0.23	*	0.10	NS	0.33
Drumsticks yield (%)	9.8	10.0	10.0	9.9	9.9	9.8	10.0	9.9	10.0	9.9	NS	0.20	***	0.09	NS	0.29
Wings yield (%)	8.0^{ab}	8.1^{a}	8.1^{a}	8.0^{ab}	8.0^{ab}	7.9^{b}	8.0^{ab}	7.9^{b}	8.0^{ab}	7.9^{b}	*	0.14	NS	0.06	NS	0.19
Remaining yield (%)	22.1^{ab}	22.1^{ab}	22.0^{b}	22.4^{a}	21.9^{b}	21.9^{b}	22.4^{a}	22.2^{ab}	22.2^{ab}	21.9^{b}	*	0.36	***	0.16	NS	0.51
Breast meat analysis[7]																
Dry matter (g/kg)	264	267	266	268	267	267	265	262	266	265	NS	4.5	**	2.0	NS	6.3
Protein (g/kg in fresh)	235	232	234	230	233	232	232	234	230	233	NS	5.8	*	2.6	NS	8.1
Fat (g/kg in fresh)	4.2	4.2	4.7	5.5	5.5	4.7	4.3	4.4	4.3	4.5	NS	1.46	NS	0.65	NS	2.07

[a–c]Significant differences ($P < 0.05$) between 2 treatment means are with different superscript letters.

[1]Data were analyzed with statistical model 1.

[2]MON 71800 is Roundup Ready wheat.

[3]MON 71900 is the parental nontransgenic cultivar.

[4]SSD = statistical significance of differences.

[5]LSD = least significant difference between 2 means.

[6]All means represent 10 pens per treatment with 9 birds per pen.

[7]All means represent 10 pens per treatment with 4 birds from each pen.

*$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

TABLE 8. Broiler performance, carcass yield, and chemical analyses of breast meat[1]

	Wheat line			Variety effect (V)	
Parameter	MON 71800	MON 71900	Reference population	SSD[2]	LSD[3]
Performance[4]					
Weight (g) d 0	43.6	42.3	42.7	NS	1.03
Weight (g) d 40	2,464	2,418	2,466	NS	44.5
Feed intake (g) 0–40 d	3,675	3,608	3,674	NS	98.4
Feed conversion 0–40 d (g/g)	1.52	1.52	1.52	NS	0.023
Carcass yield[4]					
Weight (g) d 41	2,430[a]	2,307[b]	2,408[a]	*	61.0
Slaughter yield (%)	86.8	86.7	86.8	NS	0.55
Evisceration yield (%)	87.5	87.6	87.9	NS	0.47
Breast meat yield (%)	19.6	19.4	19.8	NS	0.48
Thighs yield (%)	14.3	14.3	14.3	NS	0.36
Drumsticks yield (%)	9.8	10.0	9.9	NS	0.19
Wings yield (%)	8.0	8.1	8.0	NS	0.21
Remaining yield (%)	22.1	22.1	22.1	NS	0.52
Breast meat analysis[5]					
Dry matter (g/kg)	264	267	266	NS	4.6
Protein (g/kg in fresh)	235	232	232	NS	3.9
Fat (g/kg in fresh)	4.3	4.3	4.7	NS	1.26

[a,b]Significant differences ($P < 0.05$) between 2 treatment means are indicated by different superscript letters.

[1]Comparison of Roundup Ready wheat (MON 71800) and its parental nontransgenic control (MON 71900) with a population of 8 commercial reference wheat varieties (variety effect; mean values of males and females). Data were analyzed with statistical model 2.

[2]SSD = statistical significance of differences.

[3]LSD = least significant difference between the mean of the Roundup Ready or the control lines when compared with the mean of the reference population.

[4]All means represent 10 pens per treatment with 9 birds per pen.

[5]All means represent 10 pens per treatment with 4 birds from each pen.

*$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

TABLE 9. Broiler Performance, carcass yield, and chemical analysis in breast meat. Comparison of males and females (main effect of sex across treatments). The comparison has been made according to statistical model 2

			Sex effect (S)	
Parameter	Male broilers	Female broilers	SSD[1]	LSD[2]
Performance[3]				
Weight (g) d 0	42.8	42.7	NS	0.68
Weight (g) d 40	2,640	2,282	***	44.0
Feed intake (g) 0–40 d	3,868	3,466	***	68.9
Feed conversion 0–40 d (g/g)	1.49	1.55	***	0.005
Carcass yield[4]				
Weight (g) d 41	2,594	2,207	***	32.6
Slaughter yield (%)	86.3	87.3	***	0.21
Evisceration yield (%)	88.0	87.6	**	0.32
Breast meat yield (%)	19.4	20.0	**	0.39
Thighs yield (%)	14.4	14.3	*	0.11
Drumsticks yield (%)	10.2	9.7	***	0.11
Wings yield (%)	8.0	8.0	NS	0.09
Remaining yield (%)	21.9	22.3	***	0.16
Breast meat analysis[4]				
Dry matter (g/kg)	265	267	*	2.5
Protein (g/kg in fresh)	231	234	NS	3.1
Fat (g/kg in fresh)	4.6	4.7	NS	1.15

[1]SSD = statistical significance of differences.

[2]LSD = least significant difference between 2 means.

[3]All means represent 10 pens per treatment with 9 birds per pen.

[4]All means represent 10 pens per treatment with 4 birds from each pen.

*$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

value of Roundup Ready wheat containing event MON 71800 and used in nutritionally balanced broiler diets is comparable in performance to its parental nontransgenic control line (MON 71900) and did not differ from a population of commercially available reference wheat varieties. These results support nutritional equivalence of glyphosate-tolerant wheat with its nontransgenic counterparts in regards to performance, carcass yield, and breast meat quality.

ACKNOWLEDGMENTS

Hans Versteegh, Theo Uijttenboogaart, Henny Reimert, and Cor Pieterse of ID-Lelystad, Lelystad, The Netherlands, all contributed significantly to the planning and execution of this experiment.

REFERENCES

AOAC. 1984. Official Methods of Analysis. 14th ed. Association of Official Analytical Chemists. Arlington, VA.

Centraal Veevoederbureau. 1998. Tabellenboek Veevoeding (Dutch Feed Tables). Centraal Veevoederbureau, Lelystad, The Netherlands.

GENSTAT. 1998. Genstat5 Statistical Package Release 4.1. Lawes Agriculural Trust, IACR Rothamsted, UK.

ISO. 1996. Water quality—Determination of 33 elements by inductively coupled plasma emission spectroscopy. Method 11885. ISO, Winterthur, Switzerland.

NEN. 1974. Test methods for feedingstuffs—Determination of the starch content by enzymatic hydrolysis. NEN, Delft, The Netherlands.

Sato, H., T. Seino, T. Kobayashi, A. Murai, and Y. Yugari. 1984. Determination of tryptophan content of feed and feedstuffs by ion exchange liquid chromatography. Agric. Biol. Chem. 48:2961–2969.

Schram, E., S. Moore, and E. W. Bigwood. 1954. Chromatographic determination of cystine as cystic acid. Biochem. J. 57:35–37.

Uijttenboogaart, T. G., and A. R. Gerrits. 1982. Methods of dissection of broiler carcasses and description of parts. Report 370. Spelderholt, Beekbergen, The Netherlands.

Veerkamp, C. H., and A. H. H. Rincker. 1990. Standaard slacht en opdeelrendementen van koppels slachtkuikens. (Standard slaughter and cutting yields of broilers) Report 535. Spelderholt, Beekbergen, The Netherlands.