

## VERIFICATION OF NUTRITIVE VALUE AND HYPOCHOLESTEROLEMIC EFFECT OF SPRING BARLEY LINES IN RATS

S. Pipalová, J. Procházková, J. Ehrenbergerová, K. Vaculová

Received: May 28, 2004

### Abstract

PIPALOVÁ, S., PROCHÁZKOVÁ, J., EHRENBARGEROVÁ, J., VACULOVÁ, K.: *Verification of nutritive value and hypocholesterolemic effect of spring barley lines in rats*. Acta univ. agric. et silvic. Mendel. Brun., 2004, LII, No. 5, pp. 123-130

The aim of a growth model experiment in laboratory rats was to check the nutritive value of the set of newly bred spring barley lines with different grain characteristics. The followed set contained feed type hulless lines bred in the Agricultural Research Institute Kroměříž, Ltd. KM1771 (1), KM 1057 (2), food type hulless lines KM 2082 (3), KM 2092 (4), KM 2062 (5) and lines formed on the basis of crossing with food donors of a *waxy* endosperm type – hulless line Wabet x Washonubet (6), hulled line Wabet x Krona (7) and Wabet x Kompakt (8). The line Kompakt x Krona (9) and the malting variety Nordus were used as a control (10).

The best results of an average daily gain and feed conversion ratio (FCR) (9.19 g; 2.29) were achieved by feeding the line KM 1057 (2), conversely, the worst by feeding food barley of the *waxy* type (average daily gain in animals of experimental group 6 was only 8.01 g and FCR 2.54). This material also had the highest  $\beta$ -glucan content (7.1 %) out of the followed set. The lowest  $\beta$ -glucan content was detected in the sample 9 (3.88 %). Linear dependence between intake of  $\beta$ -glucans (x) and cholesterol content in rat's blood (y) was:  $y = -0.0591x + 3.8102$ ,  $R^2 = 0.4681$ . The found dependence was statistically highly significant ( $P < 0.01$ ).

The highest values of digestibility coefficients of crude protein (75.96 %) and crude fibre (49.63 %) were measured in animals in a group 2, on the contrary the lowest digestibility of crude fibre (31.10 %), fat (56.96 %) and ash (45.65 %) was exhibited by a control group 10. Differences in digestibility of all followed nutrients after the 14<sup>th</sup> day and at the end (28<sup>th</sup> day) of the experiment were observed (crude protein digestibility was lower by 5.65 %, ash digestibility by 11.47 %, fibre by 10.85 % and fat by 3.75 %).

It was confirmed that the tested *waxy* lines exhibited hypocholesterolemic effect; we, however, do not recommend them for feeding of monogastric animals due to high  $\beta$ -glucan content, reduced intake of mixture and impaired feed conversion.

barley,  $\beta$ -glucans, hypocholesterolemic effect, growth, rat

About 70 % of the total barley grain production is used for feeding purposes. Considerable renaissance of barley use also in human nutrition has been registered all over the world. This trend is a result of

numerous scientific and clinical research studies accentuating a health-preventive role of crude fibre consumption.

A group of structural non-starch polysaccharides

(NSP) participates in the construction of a barley grain cell wall together with lignin. The barley grain contains approximately 20 % of dietetically beneficial fibre, of which even 86 % falls on NSP, of this ca 23% are arabinoxylans and 56 % of  $\beta$ -glucans ( $\beta$ -G). These NSP belong to the most significant in nutrition of young livestock from the view of antinutritive barley effects. They swell, thicken chyme, increase its viscosity and slow down its shifting in animals' intestine and stomach (Petr, 2003). Thus mobility of nutrients and digestive enzymes is deteriorated and absorption of nutrients, mainly fats are limited (Kalač, Míka, 1997). It means that biological effects of NSP from the point of view of human nutrition, nutrition of poultry and piglets are diametrically different. While in human nutrition food fibre is appreciated favourably, first of all as one of the factors decreasing cholesterol level, in poultry and to a lesser degree in growing pigs, it reduces intake and utilizability of nutrients and utilization of barley in feeding rations of poultry or piglets without enzymatic preparations would be considerably problematic (Jeroch, Dänicke, 1995).

Obviously, barley breeding must consider the given facts and breed new varieties of various utility types with respect to the final utilization of grain.

Various forms and types of barley are used as feed barley – six-row, two-row, winter and spring, hulled and hullless forms. But the high protein content and the lowest possible  $\beta$ -G level is always required. The current feed varieties contain few essential amino acids, namely lysin, threonin, valin and izoleucin (Bhatty, Whitaker, 1987). Pursuant to Velišek (1999), lysin and leucin are the limiting amino acids. An ideotype of feed barley should therefore contain minimally 11.5 % of crude protein, but at the least 5 g of lysin/16 g of N, maximally 4.5 % of fibre and minimal content of  $\beta$ -G (Petr, Húska, 1997). Considerable increase of digestibility of an energetic grain component can be expected after elimination or reduction of a husk portion (Vaculová, 1995), therefore the question of creation of hullless barley varieties for feeding specific groups of non-ruminants is often discussed.

The spring form of barley is grown predominantly for malting purposes in Czech Republic. Crude protein content is important here too – barley workability to malt is limited by minimal (9.5 %) and maximal (11.7 %) content of crude protein and  $\beta$ -G content must be max. 4 % (Pelikán et al., 1996).

Barley grain consumption for direct food consumption is relatively low in the Czech Republic. According to the statistical data less than 30 000 tonnes, i.e. 5 % of the total production is used for these purposes annually (Vaculová, 1999). On the worldwide scale food barley is considered a very suitable material for the production of dietary products. Hypocholesterolemic and further health-preventive effect of  $\beta$ -G,

$\alpha$ -tokotrienol and active antioxidants contained in grain is verified. Varieties with increased  $\beta$ -G content (above 5 %) and higher content of digestible fibre are bred; special attention is devoted to hullless barleys and especially to barleys of the type *waxy* with the increased NSP content.

Just barley grain is prominent with its increased E vitamin content and with the highest tocotrienol proportion among cereals. On average, a barley grain contains up to 25 mg/kg E vitamin. To the vitamin isomers, both tocoferols and also tocotrienols (alfa, beta, gama and delta forms) belong. These vitamin E isomers also act as effective factors in inhibition of the key enzyme for cholesterol synthesis, of so-called hydroxymethyl-glutaryl coenzym of the A reductase (Ehrenbergerová, 2000).

Number of studies on the favourable effect of the food fibre, especially its soluble fraction, on the cholesterol content in blood serum (mainly its undesirable LDL fractions) have been published, new data with health significance, however, have been supplemented all the time (Lifschitz et al., 2002).

The object of the submitted study was to verify the nutritive value and hypocholesterolemic effect of feeding the grain of new spring barley hulled and hullless lines in a model experiment on laboratory rats.

## MATERIALS AND METHODS

Conditions of the experimental facility conform to the methodology pursuant to "Law on Protection of the Maltreated Animals" no. 246/1992 Coll.

Laboratory rat males, strain Wistar, were used as an experimental model for the growth experiment. Animals 29 days old were used in the experiment. Their average initial weight ranged within 82.1–87.1g, which complies with the standard requiring maximal differences of 5g in weight between the test groups (Kacerovský et al., 1990).

The animals were divided into 10 groups (8 experimental groups and 2 controls), and different barley material was tested in each group (assignment of materials to a number of groups is presented in Table I.). Rats were stalled in plastic cages with a slotted floor at temperature of  $23 \pm 1$  °C, air humidity 60 % and light regime 12 hours dark and 12 hours light. Experimental feed mixtures were prepared from the followed line (80 %) barley, soybean meal (16 %) and mineral and vitamin premix for laboratory rats (4 %).

Duration of the experiment was 28 days. The animals were weighed every day, body weight gains, intake feed conversion organic substance digestibility and health were followed. Residues of feeds and excrements of individual groups were separated each week, weighed, dried and analyzed for the assessment of net feed intake and apparent digestibility coefficients (%).

I: *Experimental lines and barley varieties*

Identification		Characteristics
1 <sub>B</sub> 2 <sub>B</sub>	KM 1771* KM 1057	Lines with hulless grain (of an extensive type). KM 1771 has a higher $\beta$ -G content and KM 1057 lower starch content and a higher share of essential amino acid lysine than the malting barley varieties. They were bred for <b>feed</b> utilization, line KM 1771, however, appears to be more suitable for food use.
3 <sub>B</sub> 4 <sub>B</sub> 5 <sub>B</sub>	KM 2082 KM 2092 KM 2062	Lines with hulless grain (of an intensive type). With a higher $\beta$ -G content than the registered varieties of the malting type and they are determined for <b>food</b> purposes.
6 <sub>B</sub> 7 <sub>P</sub> 8 <sub>P</sub>	Wabet x Washonubet Wanubet x Krona Wabet x Kompakt	Lines of an extensive type (created in the framework of the project GAČR-521/97/1202, solved in collaboration with DCSPB of MUAF in Brno, ARI in Kroměříž and RIBM in Brno), food type. Grains are of the type <b>waxy</b> with the increased content of proteins, $\beta$ -glucans, vitamin E and its isomers. They have an increased share of amylopectin.
9 <sub>P</sub> 10 <sub>P</sub>	Kompakt x Krona Nordus	Line Kompakt x Krona comes from crossing the varieties of a <b>malting</b> type, with intensive yield. Malting variety with a low content of proteins and $\beta$ -glucans, with intensive yield.

Notes: mark <sub>P</sub> - hulled, <sub>B</sub> - hulless barley, \* - Lines marked KM – bred in ARI Kroměříž, Ltd.

Effect of  $\beta$ -G content on changes of cholesterol levels was studied. At the beginning and at the end of the experiment, total cholesterol in blood (mmol/l) was followed in animals of the control group 9 (with the lowest  $\beta$ -G content in caryopses) and groups fed with food barley lines of the type **waxy** (groups 6, 7, 8). Blood samples from hungry animals for hematological examination of total cholesterol were taken from *v. caudalis lateralis*. The extraction point had to be well treated, because the wound was endangered by biting. The blood plasma was determined spectrophotometrically using Bio-La-Test.

Analyses of nutrient content of grain samples of experimental barley lines and experimental feed mixtures made from them were conducted based on the methods described in the notice no. 451/2000 to the law no. 244/2000 Coll. Determination of  $\beta$ -G in barley caryopses was conducted in the Research Institute of Brewing and Malting in Brno by the method Flow Injection Analysis (FIA). Content of gross energy in barley samples and in complete trial mixtures was assessed on the equipment PARR 1281 Bomb Calorimeter.

The obtained results were evaluated using the standard statistical methods (Snedecor and Cochran, 1967).

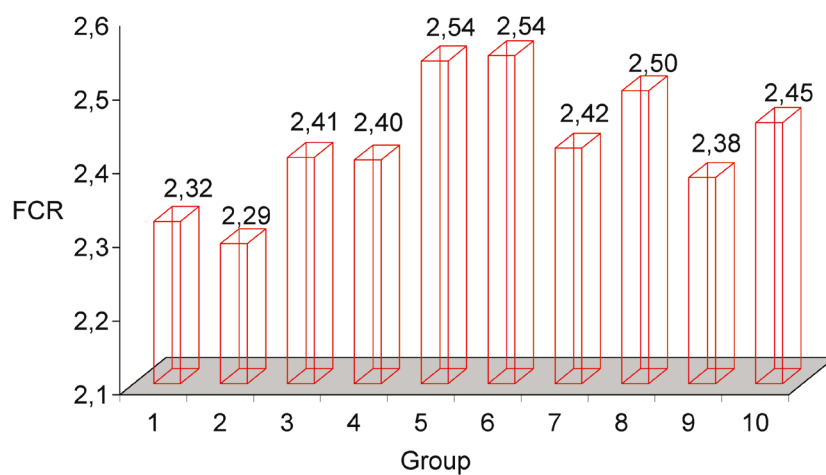
## RESULTS AND DISCUSSION

The amount of dry matter in samples varied from 88.9 to 89.4 %. The highest crude protein content was determined in the sample of feed barley 2 (151g/kg of dry matter, thereafter only DM), conversely the lowest content (118.3 g/kg of DM) was exhibited by a control group 10. The average content of crude protein was higher in hulless barleys (135 g/kg of DM) as compared with hulled barleys (127 g/kg of DM), and similarly it was higher in barley of a type **waxy** (130 g/kg of DM) compared with the standard lines and variety (133g/kg of DM). Content of organic nutrients and gross energy in grain of the followed lines and feed mixtures are given in Table II.

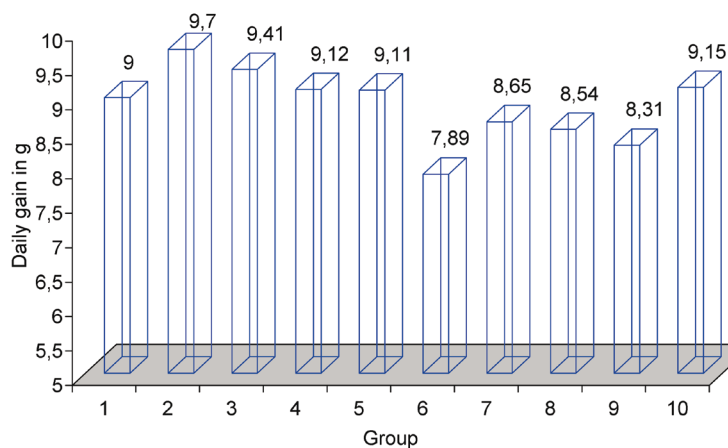
The highest average daily gain and the best feed conversion of ratio – FCR (9.70g and 2.29, respectively) were achieved by feeding the line KM 1057 (group 2). Conversely, the worst results (gain 8.01 g and FCR 2.54) were achieved by feeding food barley of the type **waxy** (group 6) – see Figure 2 and 1.

II: Content of organic nutrients, gross energy (GE),  $\beta$ -glucans ( $\beta$ -G)

Group	Crude protein (%)		Crude fibre (%)		Crude fat (%)		Ash (%)		Nitrogen-free extract (%)		GE (MJ/kg)		$\beta$ -G (%)
	Grain	Feed mixture	Grain	Feed mixture	Grain	Feed mixture	Grain	Feed mixture	Grain	Feed mixture	Grain	Feed mixture	
1	13,16	19,10	1,82	1,82	2,20	3,40	1,79	4,48	81,02	71,20	18,07	17,87	4,35
2	15,10	21,48	2,40	2,09	2,62	3,72	2,06	4,71	77,82	68,00	18,46	18,14	3,52
3	13,03	20,05	1,61	1,72	2,42	3,14	2,03	4,41	80,92	70,68	18,03	17,95	4,34
4	13,18	19,47	1,65	1,65	2,51	3,02	1,91	4,42	80,75	71,44	18,00	17,85	4,34
5	14,58	19,41	1,77	1,76	2,51	3,15	1,77	4,50	79,37	71,18	18,12	16,25	4,34
6	12,30	19,07	2,08	1,41	4,47	4,80	2,29	5,02	78,86	69,70	18,15	16,41	7,10
7	13,12	18,73	1,42	1,71	4,37	4,74	2,18	4,75	78,91	70,07	18,23	16,53	5,60
8	13,85	18,79	1,30	1,54	4,17	4,43	2,34	4,85	78,35	70,39	18,23	16,28	4,90
9	12,27	18,07	1,69	1,58	3,83	4,56	2,30	4,62	79,91	71,17	18,23	16,32	3,88
10	11,83	18,23	1,41	1,29	3,81	4,13	2,18	4,60	80,77	71,76	17,98	16,32	4,15



1: Feed conversion ratio



2: Daily gain

Apparent digestibility of crude protein, fibres, fats, ash and nitrogen-free extract of the followed utility groups in the second and the fourth week of the experiment are presented in Table III. Differences in digestibility of all followed nutrients were observed between evaluation after the 14<sup>th</sup> day and at the end of the experiment (28<sup>th</sup> day). In comparison with the second week, digestibility of all followed nutrients at the end of the experiment was significantly lower. The average values of coefficients of the apparent digestibility of the whole set indicate that the biggest drop occurred in ash digestibility (11.47 %), then in

crude fibre (10.85 %), digestibility of crude protein was lower by 5.65 % and crude fat only by 3.75 %. Concerning the course of digestibility of single utility barley types, the lowest drops were observed in groups 1 and 2, i. e. in the original feed types of barley (digestibility coefficient of crude protein was reduced only by 3.63 %, crude fibre by 4.74 %, crude fat by 2.2 % and ash by 9.66 % in comparison with the other week). Conversely, in the control groups, i.e. 9 and 10, crude protein digestibility coefficient was reduced by 10.40 %, coefficient of crude fibre even by 19.71 %, crude fat by 7.09 % and ash by 21.51 %.

III: Apparent digestibility <sup>1)</sup>

Barley	Hulles feed (average of groups 1, 2)		Hulles food (average of groups 3, 4, 5)		Food waxy (average of groups 6, 7, 8)		Standards (average of groups 9, 10)	
	week of age		week of age		week of age		week of age	
	6	8	6	8	6	8	6	8
	n = 6	n = 6	n = 9	n = 9	n = 9	n = 9	n = 6	n = 6
Crude protein	0.740 ± 2.77	0.703 ± 2.15	0.739 ± 0.59	0.700 ± 2.44	0.738 ± 1.36	0.691 ± 2.92	0.738 ± 0.55	0.691 ± 2.39
Crude fiber	0.481 ± 2.15	0.434 ± 0.33	0.437 ± 2.75	0.357 ± 1.69	0.400 ± 2.86	0.291 ± 1.53	0.400 ± 2.39	0.291 ± 2.39
Crude fat	0.640 ± 3.20	0.618 ± 3.46	0.653 ± 1.68	0.616 ± 3.31	0.650 ± 1.50	0.630 ± 3.98	0.650 ± 3.18	0.630 ± 3.53
Ash	0.494 ± 1.35	0.398 ± 0.90	0.492 ± 3.54	0.419 ± 2.70	0.495 ± 1.80	0.421 ± 2.37	0.495 ± 0.67	0.421 ± 1.02
N free extr.	0.909 ± 0.98	0.896 ± 0.33	0.916 ± 0.22	0.902 ± 0.48	0.889 ± 0.50	0.879 ± 0.43	0.889 ± 0.60	0.879 ± 1.21

1) mean ± standard error of the mean

The highest content of  $\beta$ -G in absolute dry matter of grain (Table II) was measured in the hulless food line Wabet x Washonubet - group 6 (7.1 %) and the whole group of waxy types showed significantly higher average content of  $\beta$ -G (5.8 %) versus classical lines (4.2 %).

The average content of the total cholesterol in blood of animals was 1.99 mmol/l at the beginning of the growth experiment. At the end of the experiment this value was reduced in all lines of the type waxy (in groups 6, 7 and 8 these values were 1.837, 1.981 and 1.927 mmol/l, respectively), conversely in the control group 9 (line Kompakt x Krona) the level of the total cholesterol compared to the initial value was increased to 2.83 mmol/l. Linear dependence between intake of  $\beta$ -glucans (x) and cholesterol content in rat's blood (y) was:

$y = -0.0591x + 3.8102$ ,  $R^2 = 0.4681$  (Figure 3). The found dependence was statistically highly significant ( $P < 0.01$ ).

Cholesterol content in rat's blood (mmol/l), process

of growth (total gain in g/animal/28days), intake of feed mixture and  $\beta$ -glucans (in g/animal/28 days) are summarized, presented in Table IV. The total intake of  $\beta$ -glucans and cholesterol content in rat's blood of selected groups (9 as Control group and 6, 7, 8 as experimental groups) are given in Figure 4, 5.

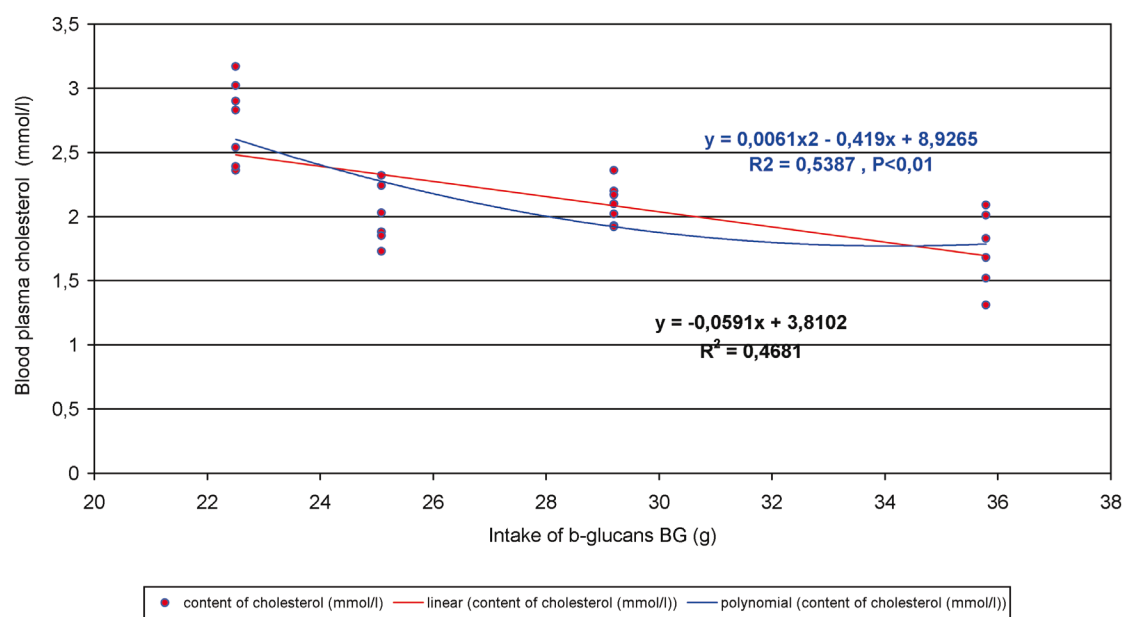
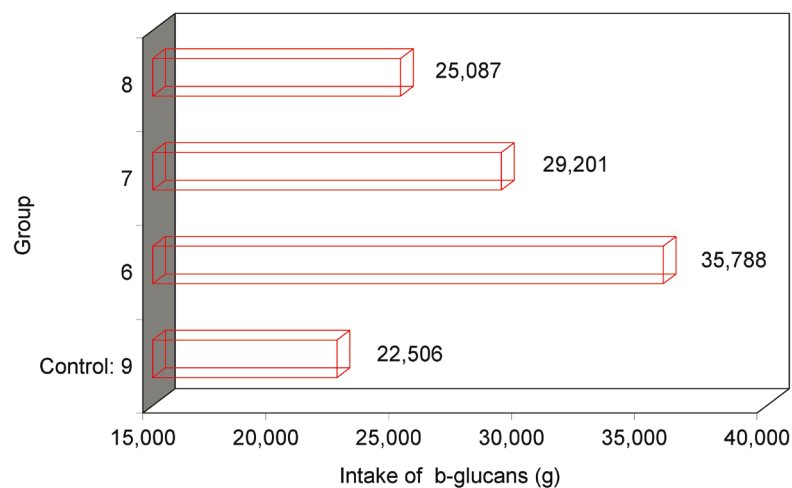
## CONCLUSION

The results suggest that the usually fed malting varieties are not the best for intensive fattening of livestock and it is necessary to look for feed barleys, preferably with hulless grain and increased content of desirable nutrients and reduced content of substances with antinutritive effect.

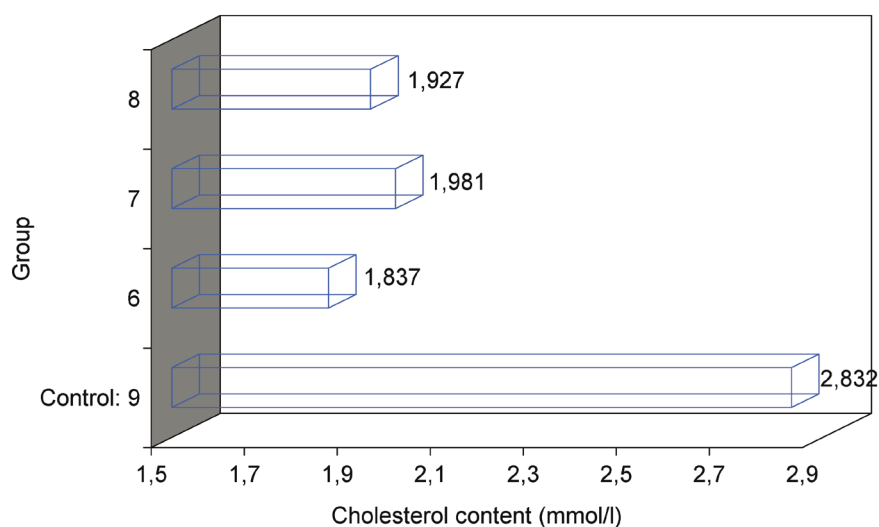
It has been confirmed that the tested waxy lines show hypocholesterolemic effect; but we do not recommend them for feeding monogastric animals due to the high  $\beta$ -glucan content, reduced intake of mixture and deteriorated feed conversion. As well as it is necessary to take their predisposition to ill health and poor yields.

IV: Cholesterol content in rat's blood, process of growth, intake of feed mixture and  $\beta$ -glucans<sup>1)</sup>

Barley	Blood plasma cholesterol concentration (mmol/l)	Total gain (g/animal/28 days)	Net feed mixture intake (g/animal/28days)	Net $\beta$ -G intake (g/animal/28 days)
	n = 8	n = 8	n = 1	n = 1
9-Control	2.832 $\pm$ 0.39	249.75 $\pm$ 31.04	517.37	22.51
6	1.837 $\pm$ 0.31	236.50 $\pm$ 45.95	504.06	35.79
7	1.981 $\pm$ 0.24	241.37 $\pm$ 16.82	521.45	29.20
8	1.927 $\pm$ 0.27	229.25 $\pm$ 36.61	511.99	25.09

1) mean  $\pm$  standard error of the mean3: The linear and polynomial dependence of reception  $\beta$ -glucans on blood plasma cholesterol concentration4: Intake of  $\beta$ -glucans





4: Cholesterol content in rat's blood

#### SOUHRN

##### Ověření nutriční hodnoty a hypocholesterolemického efektu jarních linií ječmenů na laboratorních potkanech

Cílem růstového pokusu na laboratorních potkanech bylo ověřit nutriční hodnotu souboru nově vyšlechtěných linií jarních ječmenů s rozdílnou charakteristikou zrna. Do sledovaného souboru byly zařazeny bezpluché linie krmného typu vyšlechtěné v ZVÚ Kroměříž (KM 1057 = 2, KM1771 = 1), bezpluché linie potravinářského typu: KM 2082 (3), KM 2092 (4), KM 2062 (5) a linie vytvořené na bázi křížení s potravinářskými donory waxy typu endospermu – (bezpluchá linie Wabet x Washonubet (pokusná skupina 6), pluchaté linie Wabet x Krona (7) a Wabet x Kompakt (8). Jako kontrola byla použita linie Kompakt x Krona (9) a sladovnická odrůda Nordus (10).

Nejlepších výsledků průměrného denního přírůstku i konverze krmiva (9,19g; 2,29g/g) bylo dosaženo podáváním krmné linie KM 1057 (2), naopak nejhorších zkrmováním potravinářského ječmene typu waxy (průměrný denní přírůstek zvířat pokusné skupiny 6 činil pouze 8,01g a konverze krmiva 2,54g/g). Tento materiál měl ze sledovaného souboru také nejvyšší obsah  $\beta$ -glukanů (7,1 %). Nejnižší obsah  $\beta$ -glukanů byl naměřen u vzorku 9 (3,88 %). Ze statistických údajů byla vyčíslena polynomiální závislost mezi příjmem  $\beta$ -glukanů (x) a obsahem cholesterolu v krvi potkanů:  $y = -0,0591x + 3,8102$ ,  $R^2 = 0,4681$  ( $P < 0,01$ ).

Nejvyšší hodnoty koeficientů stravitelnosti NL (75,96 %) a vlákniny (49,63 %) byly naměřeny u zvířat ve skupině 2, naopak nejnižší stravitelnost vlákniny (31,10 %), tuku (56,96 %) i popela (45,65 %) vykazovala kontrolní skupina 10. Byly pozorovány difference ve stravitelnosti všech sledovaných živin mezi hodnocením po 14. dni a na konci (28. den) pokusu (stravitelnost NL byla nižší o 5,65 %, stravitelnost popela o 11,47 %, vlákniny o 10,85 % a tuku o 3,75 %).

Potvrdilo se, že testované waxy linie vykazují hypocholesterolemický efekt; pro vysoký obsah  $\beta$ -glukanů, snížený příjem směsi a zhoršenou konverzi krmiva je však nedoporučujeme pro krmení monogastrických zvířat.

ječmen,  $\beta$ -glukany, hypocholesterolemický efekt, růst, potkan

## REFERENCES

- BHATTY, R. S., WHITAKER, J. R.: *In vivo* and *in vitro* protein digestibilities of regular and mutant barleys and their isolated protein fractions. *Cereal Chem.*, 1987, 64: 144 - 149.
- DOUBEK, J. A KOL.: *Veterinární hematologie*. 1. vyd. Brno: Noviko a. s., 2003: 464 s. ISBN 80-86542-02-5.
- EHRENBERGEROVÁ, J., VACULOVÁ, K., ZIMOLKA, J., KOUTNÁ, K.: Možnosti využití ječmene pro potravinářské účely. Nitra: Zborník z odborného seminára so zahraničnou účasťou, 1. 3. 2000, 95-99.
- JEROCH, H., DÄNICKE, S.: Barley in poultry feeding a review. *World's Poultry Sci. J.*, 1995, 51: 271 - 291.
- KACEROVSKÝ, O. a kol.: *Zkoušení a posuzování krmiv*. 1. vyd. Praha: SZN, 1990., 1050 s. ISBN 80-209-0098-5
- KALÁČ, P., MÍKA, V.: *Přirozené škodlivé látky v rostlinných krmivech*. 1. vyd. Praha: Ústav zemědělských a potravinářských informací, 1997. 317s. ISBN 80-85120-96-8.
- LIFSCHITZ C. H, GRUSAK M. A, BUTTE N. F.: Carbohydrate Digestion in Humans from a beta-Glucan-Enriched Barley Is Reduced. *J.*, 2002, 132: 2593-6.
- PELIKÁN, M., DUDÁŠ, F.: *Technologie kvasného průmyslu* – 1. vyd. Brno: MZLU, 1996, 129 s.
- PETR, J., HÚSKA, J.: *Speciální produkce rostlinná* – 1. vyd. Praha, 1997: 193. s.
- PETR, J.: O krmné jakosti ječmene. *Krmivářství*, 2003, 2: 12.
- SNEDECOR, G. W., COCHRAN, W. G.: *Statistical Methods*. 6. vyd. Iowa: State University Press, Iowa, 1967, 579 s.
- VACULOVÁ, K.: Krmná hodnota zrna obilovin pro monogastry. *Obilnářské listy*, 1995, 1: 10-12.
- VACULOVÁ, K.: Netradiční využití ječmene pro přímou lidskou výživu. *Farmář*, 1999, 10: 16-18.
- VELÍŠEK, J.: *Chemie potravin*. 1. vyd. Tábor: OSSIS, 1999, 352 s.

## Address

Ing. Sylva Pipalová, Ing. Jiřina Procházková, CSc., Ústav výživy a krmení hospodářských zvířat, Doc. Ing. Jaroslava Ehrenbergerová, CSc., Ústav pěstování a šlechtění rostlin, Mendelova zemědělská a lesnická univerzita v Brně, Zemědělská 1, 613 00 Brno, Kateřina Vaculová, Zemědělský výzkumný ústav s.r.o., Havlíčkova 2787, 767 01 Kroměříž, Česká republika