

***In situ* evaluation of the ruminal and intestinal degradability of extruded whole horsebeans**

P Cros, M Vernay*, R Moncoulon

ENSAT, Laboratoire de Zootechnie et des Productions Animales,
145, avenue de Muret, 31076 Toulouse cedex, France

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Summary — Four rumen and proximal duodenum fistulated non-lactating Holstein cows were used to determine the effect of extrusion at 120 °C of whole horsebeans (*Vicia faba* cv Talo) on *in vitro* nitrogen (N) solubility and *in situ* degradation of dry matter (DM) and crude protein (CP) in the rumen and intestine. Cows were fed a ration of 30% whole horsebeans (WHB) and 70% Italian rye-grass hay. The degradation of DM and CP was estimated using nylon bags suspended in the rumen for 2, 4, 7, 16, 24 and 48 h; the effective ruminal degradability of DM and CP was evaluated assuming a ruminal outflow rate of 0.06/h. Bags incubated in the rumen for 16 h were introduced into the small intestine through the duodenal cannula and subsequently recovered in the feces. Extrusion of WHB reduced N-solubility in buffer solution (21.1 vs 74.9%). Processing diminished the effective rumen degradability of DM (74.6 vs 80.4%) and CP (70.2 vs 89.2%). Meanwhile, the amounts of DM and CP digested in the intestine increased: 9.6 vs 1.4% and 25.2 vs 3.0% respectively. Therefore, feeds containing extruded WHB increase the availability of dietary proteins in the intestine compared with diets containing raw WHB.

horsebean / extrusion / nitrogen-solubility / *in situ* degradability

Résumé — Évaluation *in situ* des dégradabilités ruminale et intestinale de graines de féverole extrudées. Quatre vaches laitières tarées de race Holstein ont été utilisées afin de déterminer l'effet de l'extrusion à 120 °C de la graine de féverole (*Vicia faba* cv Talo) sur la solubilité des protéines et la dégradabilité *in situ* des matières sèche (MS) et azotée (MAT) dans le rumen et l'intestin. Les animaux ont reçu une ration composée de 70% de foin de ray-grass et de 30% de graine de féverole (GF). L'extrusion de GF réduit la solubilité de l'azote (21,1 vs 74,9%). Les dégradabilités ruminales de la MS et des MAT ont été déterminées par la méthode des sachets nylon, les sachets étant suspendus dans le rumen pour 2, 4, 7, 16, 24 et 48 h. Après extrusion, les dégradabilités théoriques de la MS et des MAT dans le rumen diminuent : 74,6 vs 80,4% et 70,2 vs 89,2% respectivement (avec un taux de passage théorique des particules de 0,06/h). Les sacs incubés pendant 16 h dans le rumen ont ensuite été introduits dans l'intestin par la canule duodénale et récupérés dans les fèces. L'extrusion accroît les disponibilités intestinales: 9,6 vs 1,4% pour la MS et 25,2 vs 3,0% pour les MAT. Le traitement de GF devrait donc se traduire par une augmentation de quantités de protéines d'origine alimentaire parvenant à l'intestin grêle.

féverole / extrusion / azote soluble / dégradabilité *in situ*

* Correspondence and reprints

INTRODUCTION

An adequate protein supply to high milk producing ruminants can only be assured if enough nitrogen (N) is available for rumen microbial crude protein (CP) synthesis in addition to dietary CP by-passing the rumen. Data on rumen CP degradation have revealed that not all dietary CP have the same susceptibility to microbial attack and some could be protected against digestion by treatment with heat (Ahrar and Schingoethe, 1979; Stern, 1981; Thomas *et al*, 1987; Stutts *et al*, 1988). Whole horsebeans (WHB) have attracted attention in recent years as possible home-grown protein in Europe, and are particularly high in CP and carbohydrates (Cerning-Beroard and Filiatre, 1977). Limited research has shown the benefit of heating this proteaginous seed to reduce CP breakdown in the rumen (Michalet-Doreau *et al*, 1986) and increase small intestine availability of rumen undegraded dietary CP (McMeniman and Armstrong, 1979).

The objective of our study was to determine the effect of extrusion on WHB by measuring: i), N-solubility in artificial saliva; and ii), degradation of dry matter (DM) and CP contents in nylon bags in the rumen and intestine of non-lactating Holstein cows.

MATERIALS AND METHODS

Animals and feedstuffs

Four non-lactating Holstein cows weighing 600 kg, fitted with ruminal and proximal duodenal cannulas, were kept in individual pens on a diet consisting of 70% Italian rye-grass hay and 30% of hammermilled WHB (*Vicia faba* cv Talo) 2 wk prior to and throughout the experimental period. Cows were given 50% of their assigned diet at

9.00 h and 50% at 17.00 h daily, and had free access to both feed and water. The mean daily DM intake was \approx 10 kg.

Protein sources

The CP sources tested were WHB (*Vicia Faba* cv Talo), raw (R) or extruded (E) at 120°C (Extruder Instapro, Dievet, France); EWHB was from the same batch as the RWHB. They were ground through a 1-mm screen before use in the following experiments. Nutrient composition of dietary feedstuffs is presented in table 1.

In situ incubation of protein sources

Whole ground horsebeans (5 g) were placed in duplicate bags (7 x 11 cm) of nylon cloth (Blutex T 50; Tripette et Renaud, Paris, France) with a mean pore size of 46 μ m. For incubation in the rumen, the bags were introduced immediately before the morning feedings; they were connected by a nylon cord to the cap of the ruminal cannula and kept there for 2, 4, 7, 16, 24 and 48 h. Then the bags were briefly rinsed under cold tap water to remove surface debris and machine-washed (2 x 5 min); after drying the samples were weighed.

Table 1. Dry matter (%), chemical composition (% of dry matter) of the raw and extruded whole horsebeans and the rye-grass hay.

	RWHB (10)	EWHB (10)	Rye- grass hay (10)
Dry matter	87.5	91.5	91.6
Crude protein	28.8	29.9	9.4
Ether extract	2.0	1.7	1.5
Neutral detergent fiber	23.8	22.7	68.2
Acid detergent fiber	13.9	14.3	40.4
Ash	4.7	4.9	9.2

RWHB: raw whole horsebeans; EWHB: extruded whole horsebeans; No of samples in parentheses.

The effect of extrusion of WHB on the disappearance of DM and CP in the intestine was obtained by further incubation of quadruplicate bags per feedstuff per cow. After initial incubation for 16 h in the rumen (Michalet-Doreau *et al*, 1987), the bags were removed, rinsed, incubated at 40 °C for a further 2 h with pepsin solution (3 g/l in 0.1 N HCl), rinsed again and inserted into the small intestine through the duodenal cannula (Peyraud *et al*, 1988). Bags recovered from the feces were treated similarly to those removed from the rumen. Bags not recovered within 30 h were discarded.

Biochemical analyses and calculation

Dry matter was determined in feedstuffs (105 °C overnight) and nylon bag residuals. Evaluations of fat and ash in WHB (raw and extruded), as well as in rye-grass hay were made according to the methods recommended by the AOAC (1984); neutral detergent fiber and acid detergent fiber contents of feeds were estimated by the procedure described by Van Soest (1963 a,b; 1965) and Van Soest and Wine (1967). Nitrogen (Kjeldahl) was determined both in feedstuffs and in residuals; from this, CP was determined as N x 6.25. In addition, soluble-N in each WHB protein sources was determined after extraction in artificial saliva (bicarbonate-phosphate buffer; pH = 6.9) according to the procedure of Vérité and Demarquilly (1978).

Percentage disappearance of DM and CP from the nylon bags at each incubation time was calculated from the respective amounts remaining after incubation in the rumen. Data were fitted to the non-linear regression equation: $P = a + b(1 - e^{-ct})$, where the disappearance of nutrients from the rumen, P , per time unit, t , is determined by 3 constants: a represents the rapidly soluble component, b represents the less rapidly degradable component, which disappears at the constant fractional rate, c , per time unit, t (Ørskov and McDonald, 1979). The 3 constants and fractional outflow rate per hour, k , can be used to calculate effective degradability (ED) according to the equation: $ED = a + (bc) / (c+k)$. In this study solid outflow rate, k , was not measured but an estimated value of 0.06/h was used (Vérité and Peyraud, 1988).

Ruminal breakdown of DM and CP was calculated from the proportion remaining in the bags. Intestinal digestion of DM and CP was obtained by subtraction of their ruminal degradability after 16 h of incubation from their total digestion in the digestive tract. Differences were evaluated statistically using analysis of variance. In addition, data from the rumen nylon bag study were subjected to non-linear regression analysis and the constants, a , b , and c were fitted by an iterative least squares procedure. Results are presented as means and standard errors.

RESULTS AND DISCUSSION

Extrusion of WHB was followed by an effective reduction of the N-solubility in artificial saliva (21.1 vs 74.9%); this finding is in agreement with those of others on the effect of heat treatment on N-solubility (Schingoethe and Ahrar, 1979; Michalet-Doreau *et al*, 1986; Leonard and Block, 1988).

Because the retention time of feedstuffs in the rumen is, among others, a function of amount fed, the effect of extrusion on the digestion of WHB in the rumen was examined for different ruminal incubation periods (fig 1). After 2 h of incubation, the disappearance of DM and CP from the rumen was significantly higher ($P < 0.001$) for RWHB than for EWHB. However, with longer incubations, the effect of extrusion gradually decreased; thus after 48 h in the rumen, there was no significant reduction in disappearance of DM and CP for EWHB compared to the untreated grains.

The parameters defining the equations describing the pattern of degradation of DM and CP as well as the EDDM and EDCP values are presented in table II. Coefficients of determination (r^2) for disappearance of DM and CP were significant ($P < 0.001$). For RWHB, the amount of rapidly soluble-N (a) in the rumen was high

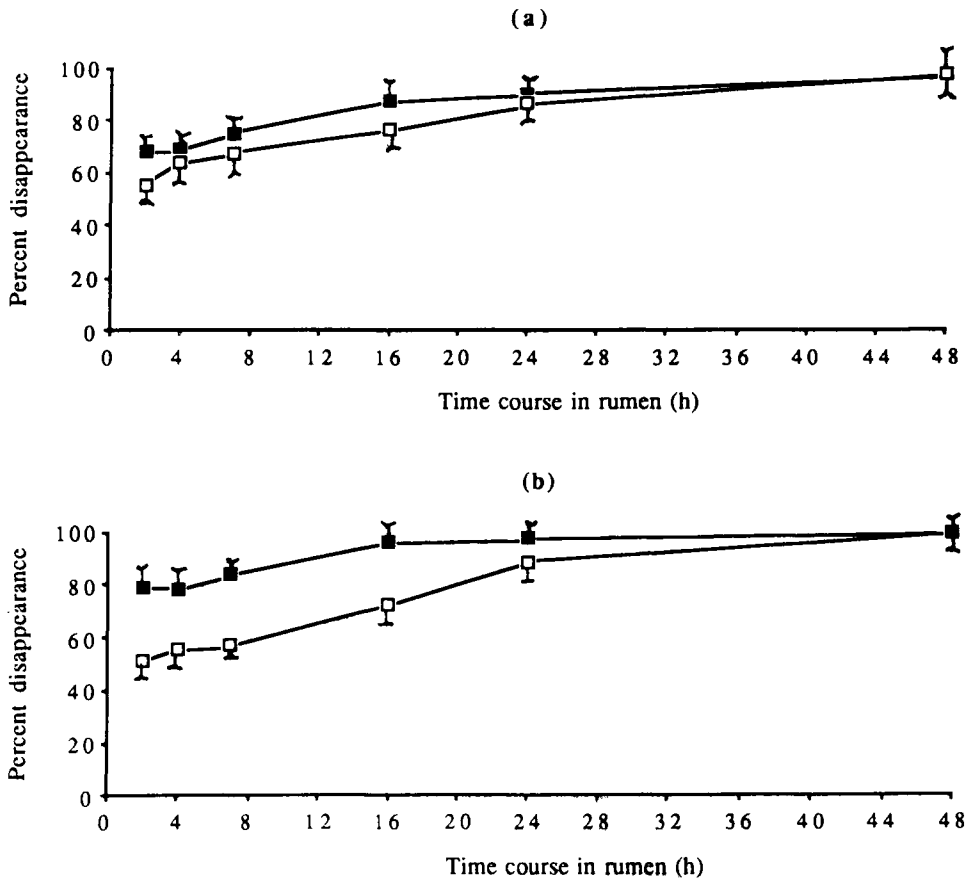


Fig 1. Effect of extrusion on percentage disappearance of dry matter (a) and crude protein (b) of whole horsebeans, raw (■) or extruded (□), from nylon bags as a function of ruminal incubation time. Each vertical bar indicates the standard error of difference between 8 means.

(67.9%), although in agreement with buffer solubility. Likewise, Michalet-Doreau *et al* (1986) observed a high value of this constant in the N-disappearance study for the *Vicia faba* cv Ascott (76.3%) and a lower value (59.8%) for *Vicia faba* cv Alto. The EDCP value for RWHB (89.2%) agrees well with values obtained previously by others (Michalet-Doreau *et al*, 1986; V \acute{e} rit \acute{e} and Peyraud, 1988) for the same legume seeds. Although, the general effects of

treating WHB on non-linear regression parameters of DM and CP were similar, some differences were noted in the response to extrusion. As expected, treatment of WHB was followed by a decrease in the proportion of the rapidly degraded DM and CP fractions, *a*, and both increased the proportion and reduced the breakdown rate, *c*, of the fraction *b*, for the WHB. Consequently, EDDM and EDCP were reduced by 7.2 and 21.3% respectively.

Table II. Effect of extrusion on non-linear parameters and effective degradability of dry matter and crude protein of the whole horsebeans.

	Dry matter		Crude protein	
	RWHB (48)	EWHB (48)	RWHB (48)	EWHB (48)
a (%)	60.8	53.2	67.9	38.5
b (%)	36.2	44.4	31.1	60.6
c (%)	7.1	5.6	13.1	6.6
r ²	0.95	0.90	0.94	0.84
ED (%)	80.4	74.6	89.2	70.2

RWHB: raw whole horsebeans; EWHB: extruded whole horsebeans; number of samples in parentheses. a, b, c = non linear parameters; ED = effective degradability (see p 251); r² = coefficient of determination.

Total disappearance of DM and CP from the digestive tract is depicted in figure 2. The percentages observed for the whole tract digestion of DM (85.8–88.5%) and CP (97.2–98.7%) were high, but approximately similar to those obtained by de Boer *et al* (1987), with soybean meal, canola meal and fishmeal and by Arieli *et al* (1989) with

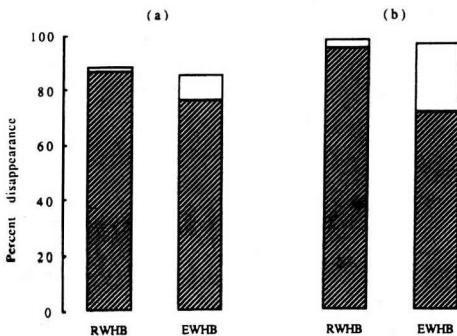


Fig 2. Effect of extrusion on relative proportions of ruminal (▨) and intestinal (□) disappearance of dry matter (a) and crude protein (b) from whole horsebeans. RWHB: raw whole horsebeans; EWHB: extruded whole horsebeans.

whole cottonseeds. Processing of WHB significantly ($P < 0.001$) depressed DM and CP digestion during rumen exposure for 16 h, but the whole tract disappearance of these components was not significantly affected. Therefore, extrusion of WHB augmented the amount of DM and CP digested in the intestine (9.6 vs 1.4% and 25.2 vs 3.0%, respectively); these results are in accordance with the linear increase in the flow of CP to the duodenum from soybean meal (Plegge *et al*, 1985), whole soybeans (Stern *et al*, 1985) and whole cottonseeds (Pena *et al*, 1986) heated from 115 to 149 °C. Nevertheless, *in vivo* studies (McMeniman and Armstrong, 1979), evaluating the incidences of heat-treatment of WHB on intestinal CP availability, have yielded different results, *eg* processing did not significantly enhance the flow of total-N in the duodenum. The reason for this discrepancy between *in vivo* and *in situ* studies can be attributed to the temperature reached by the WHB (105 °C). Indeed, this view is supported by observations for rapeseed (Lindberg, 1984) and soybean (Mir *et al*, 1984) meals as well as for canola and soybeans (Deacon *et al*, 1988), where moderate heating of the meals and seeds did not have a pronounced effect on ruminal degradation.

Whole horsebeans CP were effectively protected from degradation in the rumen by extrusion at 120 °C without adverse effect on CP total degradability. The increase in the supply of dietary CP to the post-ruminal sections as a result of extrusion could be of rapid benefit to high yield cows. Appropriate trials with these animals are now in progress.

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