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## Characterization of protein fractions of sunflower seeds during rumen degradation

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Cette étude intra-matière première montre qu'il est possible de prédire la dégradabilité ruminale de l'azote du *corn gluten feed* avec une précision suffisante en utilisant à la fois des critères chimiques [cellulose brute, action du pH sans les enzymes (TE2)] et des critères biologiques [dégradabilité enzymatique (DE1)].

La solubilité enzymatique à pH ruminal (DE2) ne semble pas mieux prédire la DT6 que la solubilité par action du pH (TE2). Ceci montre que les *corn gluten feed* testés ont subi un traitement thermique modéré et que leurs protéines sont facilement accessibles. La prédiction de la dégradabilité des matières azotées totales des *corn gluten feed* par TE2 et la cellulose brute [2] présente l'intérêt d'être la plus précise et la moins coûteuse.

**Characterization of protein fractions of sunflower seeds during rumen degradation.** R Nola <sup>1</sup>, S lametti <sup>2</sup>, F Sessa <sup>2</sup>, GF Greppi <sup>3</sup>, S Pagani <sup>2</sup> (<sup>1</sup> *Ist Zootec Gen, Milan*; <sup>2</sup> *Dip Sc Mol Agr Alim, Milan*; <sup>3</sup> *Dipartimento di Scienze Anatomiche, Fisiologiche e delle produzioni animali, viale della Piaggera, 2, 56121 Pisa, Italy*)

The protein value of a feed utilized in polygastric nutrition depends on rumen degradation as well as on the amino-acid composition and digestibility of the undegraded proteins passing to the small intestine. The nylon-bag method is an effective tool for the prediction of rumen degradation, but further indications can be provided by investigating the rumen degradability of specific protein fractions. In this study we monitored the breakdown rates of sunflower-seed protein fractions during rumen incubation. The amino-acid composition of slowly degradable fractions was determined with the aim of correlating these polypeptide fractions with the level of microbial proteolysis in the goat rumen and the post-ruminal amino-acid availability.

Sunflower seeds were incubated in goat rumen (0, 2, 4, 8, 12, 16, 24, 48 and 72 h) and the protein fraction of the undegraded residues was solubilized. Samples from each ruminal incubation time and containing 20 µg of nitrogen were electrophoresed on 15% polyacrylamide gel according to Laemmli. Fractional protein degradation was monitored by image processing. The slowly degradable polypeptides were transferred from

the electrophoretic gel to a polyvinylidene difluoride membrane and directly hydrolysed for amino-acid analysis.

Amino-acid analysis was performed by reversed-phase HPLC (C18) after derivatization with phenylthiocyanate. The protein fractions of apparent  $M_r$  of 21, 23, 32, 34, 42 and 57 kDa showed different time-courses of degradability. None of these bands were detected by SDS-PAGE after 48 h rumen incubation. The 21 and 42 kDa polypeptides showed the major resistance to rumen microbial proteolysis with respective degradation percentages of 3.6 and 24.5 after 2 h, 74.4 and 54.4 after 8 h, and 86.6 and 91.2 after 24 h.

The relative amino-acid distributions of the 21 and 42 kDa were the following: 30.4 and 22.4 hydrophobic residues, 0.6 and 2.3 cysteine, and 14.0 and 14.7 basic residues, respectively.

Our results show that the study of fractional protein degradation is an important task in order to find dietary protein fractions with higher rumen bypass. The amino-acid composition of sunflower seed 21 and 42 kDa polypeptides indicated that slight amino-acid differences can be responsible for different susceptibilities to rumen microbial proteolysis.

**Effect of the rate of energy supply on nitrogen digestion in dairy cows fed a fresh forage diet.** JL Peyraud, L Astigarraga (*Station de Recherches sur la Vache Laitière, 35590 Saint-Gilles, France*)

The digestion of high-nitrogen (N) grass diets is characterized by a high rate of ammonia production and ruminal N losses. N could be used more efficiently if the rate of dietary energy supply and N release were synchronised.

The effects of supplementing white clover (42.5 gN/kg DM) with wheat, whose rate of energy release matches ammonia production from white clover, or beet pulp (respectively 93 and 51% digestible OM (DOM) disappearing after 4 h incubation *in sacco*), on digestion were investigated in a cross-over design on 6 fistulated dairy cows (21 kg milk). Fresh grass was given *ad libitum* in 3 meals per day. Concentrates made up 30% of the diet and were distributed before the first and the last forage meals. Wheat straw (500 g/d) was offered to prevent bloating. The flow of duode-