Biofuel by-product for poultry diets (ruminal cellulosome)

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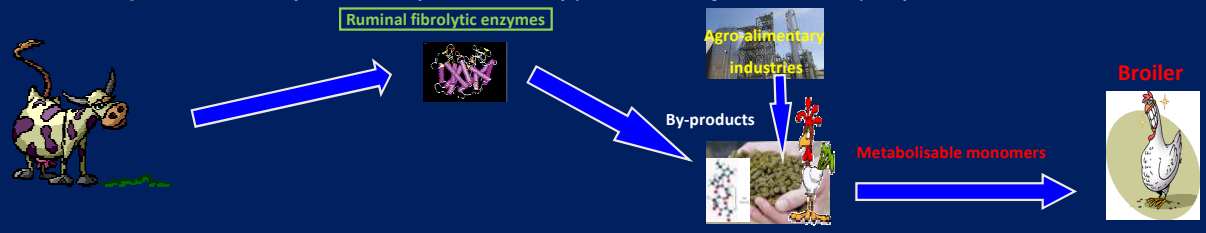
INTRODUCTION : The valorization of by-products from biofuel industry will promote the application of the 2010 (2003/30/EC) and 2020 European Directives, stipulating the inclusion of biofuels in transport sector. Replacement of fossil energy for sustainable energy by cereal utilization in bioethanol production increases the competition for starch between monogastric organisms and first generation biofuels. Very wide and interesting opportunities toward Sustainable Development are opened by by-product valorization.

The aim of the study : Utilization of fibrolytic ruminal enzymes to valorize by-products in the digestive tract of the poultry.



CRUDE EXTRACT FROM RUMINAL SOLID CONTENT

Fiber adherent bacteria (cellulosome) = 80-90 % of ruminal cellulolytic activities

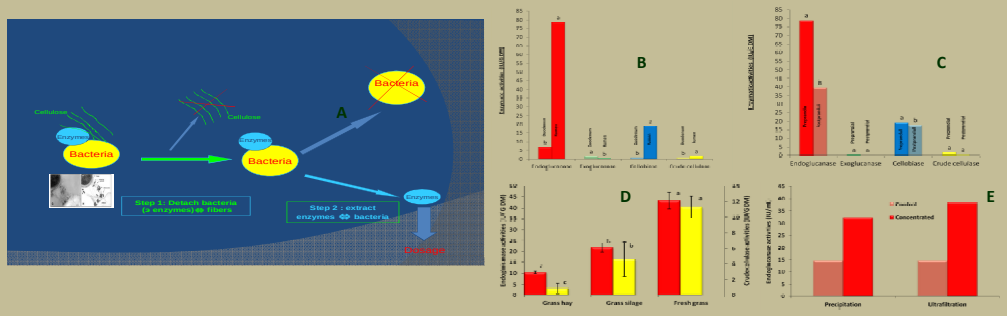


Figure 1: General methodology to produce crude extract from ruminal solid content (A: Cellulosome structure G-H ; Bayer et al., *2004*) and conditions of crude extract production (B: **ruminal activities** *vs* duodenal activities, C: **preprandial** *vs* postprandial, D: grass hay *vs* grass silage *vs* **fresh grass,** E: activities concentration by precipitation ***vs* ultrafiltration**)

CELLULOLYTIC POTENTIAL OF RUMINAL CRUDE EXTRACT ON BY-PRODUCTS

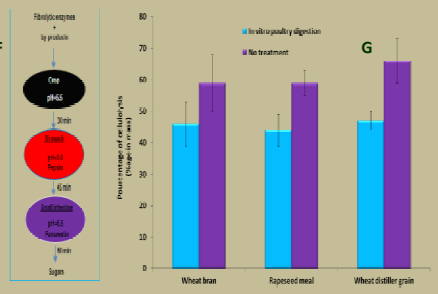


Figure 2: Hydrolysis of cellulose content (G: 44-66% depends on the by-products tested) with and without *in vitro* digestion simulation of poultry (F: pepsin, pancreatin and stomach acidity).

ISOLEMENT OF CELLULOLYTIC RUMINAL BACTERIA

Different technics were used to isolate cellulolytic bacteria from ruminal microorganism consortium.

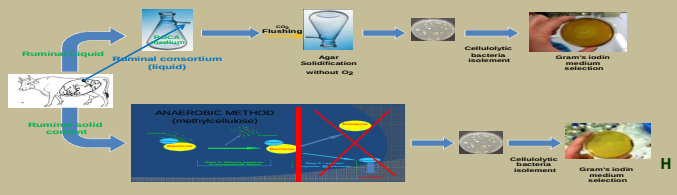


Figure 3 : Hydrolysis halo from cellulolytic ruminal bacteria cultivated and isolated anaerobicaly on specific medium. Three strains were selected for their cellulolysis potentialities and were identified by 16S rDNA sequencing. Identification revealed a recently isolated strain from lignocellulose : *Bacillus niabensis* (H: Kwon et al., 2007).

FIBROLYTIC BACTERIA PRODUCTION FROM *EX VIVO* SYSTEM

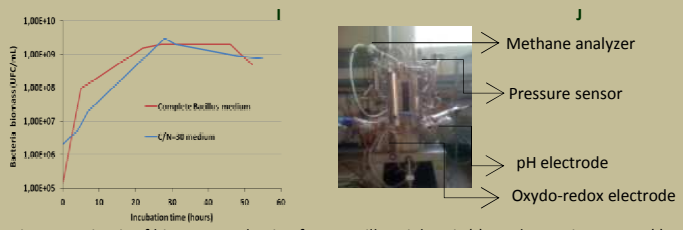


Figure 4: Kinetic of biomass production from *Bacillus niabensis* (J). In the *ex vivo* system (I), the cellular biomass reached 10^9 bacteria/mL from 10^5 bacteria/mL in 24 hours (10 litter bioreactor).

FIBROLYTIC ENZYME PRODUCTION FROM *EX VIVO* SYSTEM

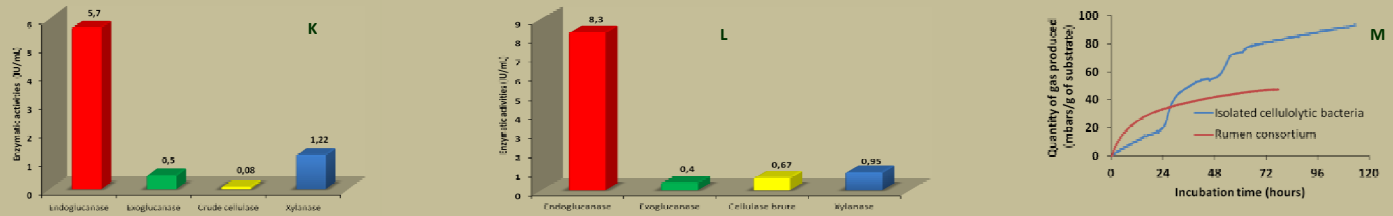


Figure 5: Fibrolytic enzyme production after 24 hour culture on C/N=30 medium from rumen cellulolytic bacteria isolated from ruminal liquid (K) and solid (L: fiber adherent bacteria). These enzyme productions from *Bacillus niabensis* are both hemicellulolytic and cellulolytic (reached 5,7 IU/mL of endoglucanase activities in the *ex vivo* system). Strains are facultative anaerobic. The anaerobic potentialities of their growth are investigated by gas production in order to control and to caraterize the cellulolytic fermentation on different cellulosic substrates (carboxymethylcellulose, avicel and distilled dried grains with solubles); gas production curves (M) show the difference between anaerobic ruminal consortium and isolated cellulolytic strain.

Conclusion : Ruminal fibrolytic enzymes (crude extract) showed *in vitro* potentialities on cellulosic substrates and hydrolyzed 44 to 66% of distilled dried grains with solubles (wheat distiller grains with and without monogastric digestion simulation). Cellulolytic strains were isolated from ruminal content and cultivated in the *ex vivo* system (C/N=30 medium). Production of fibrolytic enzymes in fermentors by bacteria culture stimulation permitted to obtain the rate of activity needed to make *in vivo* experimentation with monogastric animals.

Bibliography: Bayer et al., THE CELLULOSOMES Multienzyme Machines for Degradation of Plant Cell Wall Polysaccharides. Annulal review of microbiology (2004) Vol 58: 521-554. Kwon et al., *Bacillus niabensis* sp.nov., isolated from cotton-waste compost for mushroom cultivation. International Journal of systematic and Evolutionary Microbiology (2007) Vol 57: 1909-1913.

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