# Effects of an additive enriched with the first limiting amino acids on growing performances of double-muscled Belgian Blue bulls fed a corn silage based diet

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#### Abstract

Two trials were conducted on double-muscled Belgian Blue bulls (BBb). The aims of the first one were (1) to determine the ruminal bypass of free amino acids (Met and Lys) administred intraruminally (once per day during the morning meal) and (2) to determine the lack of the first limiting amino acids (AA) of BBb for a corn silage based diet in order to formulate a specific additive that was investigated in the second trial. The first experiment was led on 6 fistulated animals according a 6x3 cross-over design. The results suggested that His, Met, Arg, Lys and Val were respectively the first limiting AA in our experimental conditions. The mean ruminal escape of Met and Lys was similar and particularly high (bypass = 37%) due to the low volume of rumen liquid and the high particle outflow rate. The second experiment was led on 24 animals in a randomised blocks design. The additive had only a significant effect on growth performances during the growing period of the BBb (370–430 kg) with an increase of ADG by 255 g/d.

#### Introduction

The Belgian Blue is the main beef breed used for meat production in Belgium. Due to their high growth potential, the Belgian Blue bull (BBb) has specifics amino acids (AA) requirements (Froidmont et al., 2000) that are not completely satisfy with practical diets. As shown by a previous study (Froidmont et al., 2002), dietary incorporation of free AA can be a useful way to improve the AA pattern of metabolisable protein in order to satisfy the AA requirements of BBb. Two studies were conducted to investigate the opportunity to supply free AA in BBb fed with a corn silage based diet. The aims of the first one were (1) to determine the ruminal bypass of free amino acids (Met and Lys) administred intraruminally (once per day during the morning meal) and (2) to determine the lack of the first limiting amino acids (AA) of BBb for a corn silage based diet in order to formulate a specific additive that was investigated in the second trial.

#### Material and methods

## <u>Trial 1:</u>

Three treatments were tested according to a 6 x 3 duplicated Latin square design experiment on six double-muscled Belgian Blue bulls  $(337 \pm 22 \text{ kg})$  fed with the same diet (table 1) at an intake level of 85 g/kg<sup>0.75</sup>. After 21 d of diet adaptation, each experimental period was composed by 2 d for treatment transition, 9 d for urine and feces collection, 2 d for duodenal and ileal contents sampling, and 1 d for ruminal fluid collection. The treatments were administered in order to supply in the rumen 0 g of DL-Met and 0 g of L-Lys [Control treatment], 40 g of DL-Met and 60 g of L-Lys administered once per day at the morning meal [Flash treatment] or 40 g of DL-Met and 60 g of L-Lys infused throughout the day [Continuous treatment]. These AA were dissolved in 500 mL of water containing 1.4 g of Co as CoEDTA, used as a liquid phase tracer. Chromic sesquioxyde (10 g/meal, in the rumen) was used as DM tracer. The by-pass of AA was determinated according to the protocol described by Cottle and Velle (1989).

## Trial 2:

Twenty-four BBb  $(331 \pm 26 \text{ kg})$  were randomly arranged in 4 pens (6 animals per pen) and received 4 dietary treatments (2 diets ["normal" *vs* "low" in protein] x 2 treatments [with or without additive]) (table 1). The additive (300 g/bull), including 86.5 g free AA (His [28.0 g], Met [10.0 g], Arg [20.4 g], Lys [20.5 g] and Val [7.2 g]) and 45 g dextrose mixed with cereal bran, was distributed directly on the meal. Bulls were weighed monthly throughout the trial and the daily intakes were recorded individually. The trial was divided into 3 periods (phase I : 0-42 d, phase II : 43-104 d, phase III : 104-169 d) for better measuring the additive effect at different stages of growth.

	Trial 1	Trial 2			
g/kg DM	Diet	« Normal » diet	« Low » diet		
Ingredients					
Corn silage	489	499	700		
Soya cake	200	190	60		
Dry beet pulp	193	235	181		
Hay	100	61	16		
Minerals and vitamins	18	15	20		
Protected oil	-	-	17		
Urea	-	-	6		
Nutritional value <sup>1</sup>					
Crude protein	162	168	106		
VEVI	995	1000	1000		
DVE	103	106	70		
OEB	12	8	-4		

Table 1. Composition and nutritional value of diets in trials 1 and 2

<sup>1</sup>According to Dutch feeding standard, VEVI = net energy; DVE = digestible protein in the small intestine; OEB = difference amongst rumen microbial protein synthetised with available N and with available energy.

#### **Results and discussion**

## <u>Trial 1</u>

To display the limiting AA of BBb fed with a corn silage based diet, we proceeded in two steps. The first one consisted to determine the digestible AA flows in bulls receiving such diet (table 2). The second one was to establish the requirement of each essential amino acid (EAA). The EAA requirements for the BBb were estimated by proportioning the requirements in Met and Phe, determinate previously by Froidmont et al. (2000), to the AA profile of the carcass (De Campeneere et al. 2001). In our experimental conditions, the comparison of digestible EAA supply with the animal's requirements showed that the limiting AA are respectively histidine, methionine, arginine, lysine, valine, threonine, leucine, tryptophane and phenylalanine (table 2). Met and Lys were generally the most frequently limiting AA recorded in the literature but others EAA, such as histidine, can also be limiting (Greenwood and Tigemeyer, 2000; Schoof et al., 2000; Wessel et al., 1997).

Table 2. EAA classification according to their deficiency

EAA (g/d)	His	Met	Arg	Lys	Val	Thr	Leu	Trp	Phe
Digestible EAA supply (g/d)	12.5	10.5	37.0	38.8	28.3	25.8	48.9	2.8	29.7
Digestible EAA requirement (g/d)	34.9	22.5	71.9	75.0	51.1	42.5	79.1	4.3	41.4
Supply / requirement (%)	35.9	46.7	51.5	51.8	55.5	60.7	61.8	62.6	71.8

On the basis of ruminal escape of Met and Lys, the "Flash" treatment induced a higher AA bypass (37%) than the "Continuous" treatment (15%). The high bypass value with the "flash" treatment was due to the low volume of rumen liquid (35 litters) and the high particle outflow rate (15%/h). This value was similar of that observed by Froidmont et al. (2002).

N retention was not significantly different between treatments ("Control":  $54,0 \pm 11,5 -$  "Flash":  $55,4 \pm 11,7$  and "Continuous":  $55,3 \pm 11,4$  gN/d), probably because His was more limiting than Met and Lys.

The additive was formulated by considering a free AA outflow of 30% and for satisfying a minimum of 60% of each EAA requirement, implying to include the first five limiting AA. It was supplied once a day at the morning meal. This choice was made to find a good compromise between the requirements satisfaction, the efficiency of digestible EAA use and the cost of additive production. The additive was composed of 86.5 g free AA (His [28.0 g], Met [10.0 g], Arg [20.4 g], Lys [20.5 g] and Val [7.2 g]) and 45 g dextrose mixed with cereal bran.

# Trial 2

The additive effect was significant only during the first experimental phase (table 3) with an increase of the average daily gain (ADG) of 255 g (+ 21%) and a reduction of food conversion ratio by 1.8 (-25%), compared to the bulls who did not receive the additive. These positive effects were more effective with the "low" protein diet where a supplemental ADG of 357 g was recorded (*vs* 155 g for the "normal" diet). These results confirm that large amount of free AA can escape from ruminal degradation (Froidmont et al., 2002) and allow an improvement of the N valorization as much better that the crude protein content of the diet is low (33.7 *vs* 23.8 g N retained/100 g N ingested for "normal" diet).

Criteria	Ad	lditive	SEM	p value	
	without	with			
ADG (kg/d)					
Phase I*	1.199	1.454	0.058	0.040	
Phase II*	1.185	1.155	0.039	0.703	
Phase III*	1.105	1.177	0.035	0.316	
FCR					
Phase I*	6.92	5.13	0.401	0.037	
Phase II*	7.05	7.16	0.232	0.820	
Phase III*	8.17	7.56	0.212	0.163	

Table 3. Additive effect on average daily gain (ADG) and food conversion ratio (FCR)

\*Phase 1 : 370-430 kg live weight (LW) ; phase 2 : 430-500 kg LW ; phase 3 : 500-570 kg LW

In the following phases (II and III), the additive did not have any more effects on ADG or FCR. We supposed that the quantity of distributed additive, who was the same during whole trial, appeared insufficient beyond 450 kg of live weight. However, at this physiological stage, energy can also be limiting, decreasing by this way the efficiency of AA use.

#### Conclusions

The results of the first trial confirmed that a significant part of AA distributed in a free form can escape from ruminal fermentations, provided that these AA leave the rumen quickly. However, the supplement of methionine (Met) and lysine (Lys) distributed to animals did not have significant effects on zootechnical performances, probably because histidine (His) was the first limiting AA.

The second trial showed that the use of the additive enriched with the first limiting AA in a free form increased the daily gain and improved the food conversion ratio by bulls in their optimal growth phase (between 370 and 430 kg) receiving a corn silage basal diet. These results should be confirmed on lighter animals (250 kg), when the quality of digestible protein determines even further the valorization of the diet.

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