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EFFECT OF CELLOBIOSE SUPPLEMENTATION IN DRINKING WATER AND FEED RESTRICTION ON APPARENT FAECAL DIGESTIBILITY AND GROWTH PERFORMANCE IN RABBITS

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How to cite this paper
EFFECT OF CELLOBIOSE SUPPLEMENTATION IN DRINKING WATER AND FEED RESTRICTION ON APPARENT FAECAL DIGESTIBILITY AND GROWTH PERFORMANCE IN RABBITS

Farias-Kovac C., Simbaña F., Reyes M., Ávila A.B., Carabaño R., Nicodemus N., García J.*

Dpto. Producción Agraria, Universidad Politécnica de Madrid, Paseo Senda del Rey 18, 28040, Madrid, Spain
*Corresponding author: javier.garcia@upm.es

ABSTRACT

The objective of this work was to evaluate the effect of cellobiose supplementation in water (CEL) and its potential synergy with feed restriction on faecal digestibility and growth performance. Four treatments in a factorial arrangement were used: 2 levels of CEL (0.0 and 7.5 g/L) × 2 feeding plans (ad libitum and restricted, from 32 to 47 d of age). A total of 236 32-d old rabbits weighing 700 ± 116 g were blocked by litter randomly assigned to the four treatments and caged individually until 60 d of age. The restricted group was fed with 50% of the feed eaten by the ad libitum group at weaning and the daily feed supply increased linearly until 100% of intake of the ad libitum group at 47 d of age. Fecal digestibility was determined between 39 and 43 d (D1) and between 53 and 56 d of age (D2) (10/treatment). Cellobiose supplementation had no effect on faecal digestibility but tended to increase starch digestibility in D1 (P = 0.074). Feed restriction improved energy, protein (both by 5%), starch (+0.3%) and total dietary fibre digestibility by 11% (P ≤ 0.026) in D1, with no effect in D2. Feed efficiency improved in the whole experimental period with cellobiose supplementation (+3%. P = 0.003), due to the trend to increase the growth rate (P = 0.11), with no effect on feed intake and mortality. During the restriction period feed intake of restricted rabbits was a 72% of that of the ad libitum group, while in the whole experimental period accounted for a 90% of the ad libitum group. As expected, it decreased growth rate (-3.5%; P = 0.015) and improved feed efficiency (+7%. P < 0.001) in the whole period, resulting in a lower final liveweight (2287 vs. 2231 d g; P = 0.015). Feed restriction, tended to reduce mortality rate (18.6 vs. 10.1%; P = 0.067), and curiously no differences in mortality were observed during the restriction period, and this tendency was explained by reduction of the mortality during the refeeding period (P = 0.041). There was no relevant interaction between CEL and feed restriction. In conclusion, CEL supplementation improved growth traits but had no influence on mortality, while gradual feed restriction, tended to decrease mortality with a minor reduction of growth traits.

Keywords: Cellobiose, Feed restriction, Digestibility, Growth performance, Rabbit.

INTRODUCTION

Cellobiose supplementation in drinking water (CEL, 7-7.5 g/L) reduced mortality rate when it was combined with a low soluble fibre diet (8.4%, on DM basis) in a context of epizootic rabbit enteropathy, but a higher dose (15 g/L) increased mortality (Ocasio-Vega et al., 2018a, 2019). The positive effect of cellobiose might be related to the increase of the proportion of butyrate in the ileal digesta, effect also observed in the caecal in vitro fermentation (Ocasio-Vega et al., 2018a,b). Feed restriction programs have proven to be effective in reducing mortality and optimizing feed efficiency after weaning in rabbits, although has the disadvantage of the impairment of growth traits when the...
fattening period is short like in Spain (Romero et al., 2010; Gidenne et al., 2012). An alternative might be a progressive feed restriction plan (Duperray and Guyonvarch, 2009; Birolo et al., 2016), that helps to control mortality but still impaired growth traits (Farias-Kovac, 2021). The aim of this study is to evaluate the possible synergic effect of cellobiose supplementation with feed restriction on faecal digestibility and growth traits.

MATERIALS AND METHODS

Experimental design
Four treatments in a 2 x 2 factorial arrangement were used with two levels of CEL (0, and 7.5 g/L. Savanna Ingredients GmbH, Elsdorf, Germany) along the whole fattening period, and two feeding plans (ad libitum, AL; restricted, Rest.). Restriction started with a 50% of the AL group and increased progressively until 100% at 47 d of age inspired in the feeding plan studied by Duperray and Guyonvarch (2009) and Birolo et al. (2016). From 47 to 51 d restricted rabbits were offered the same feed eaten by the AL group, and from 51 to 60 d they were fed ad libitum. A control diet was formulated to meet the nutrient requirements for growing rabbits with 21.1% crude protein, 35.5% neutral detergent fibre, 9.1% soluble fibre, 13.4% starch and 19.0 MJ/kg gross energy (on DM basis). A total of 236 rabbits weighing 700 ± 116 g were weaned at 32 d of age, blocked by litter, randomly assigned to the four treatments and individually caged. Due to the design of the farm, treatments were not balanced (AL-CEL− =56 rabbits, R-CEL− =56, AL-CEL+ =62, R-CEL+ =62). No antibiotic was supplied. Rabbits had ad libitum access to water. Faecal digestibility was determined (9/treatment) from 39 to 43 d age (D1), and from 53 to 56 d of age (D2). Growth traits were recorded at 32, 47 and 60 d of age.

Statistical Analysis
Data were analyzed as a completely randomized block design with level of CEL, feed restriction and their interaction as the main sources of variation and litter as a block, weaning weight was used as a covariate for growth traits by using a mixed model. Mortality was analyzed using a logistic regression considering a binomial distribution including the same variables in the model, and the results were transformed from the logit scale. All data were presented as least-squares means.

Chemical Analyses
Procedures of the AOAC (2000) were used to determine DM (method 934.01), crude protein (968.06), starch (amylolucosidase-α-amylase method; method 996.11), and total dietary fibre (985.29), with no mucin correction for faeces. Gross energy was measured by adiabatic bomb.

RESULTS AND DISCUSSION
Cellobiose supplementation had no effect on faecal digestibility (D1 and D2), but a trend to improve starch digestibility in D1 (P = 0.074. Table 1). The lack of effect on the total dietary fibre digestibility contrast with the modification of in vivo and in vitro fermentation observed previously with CEL supplementation (Ocasio-Vega et al., 2018a,b). Cellobiose supplementation improved feed efficiency in the whole experimental period by 3% (0.404 vs. 0.416; P = 0.003), due to the trend to increase the growth rate (P = 0.11. Table 2) with no effect on feed intake, which is similar to the results obtained by Ocasio-Vega et al. (2018a, 2019). Nevertheless, these authors reported a positive influence of CEL supplementation (combined with a low dietary soluble fibre level) on mortality, but in this study no effect of CEL on mortality was observed. Feed restriction improved in D1 faecal digestibility of energy (63.8 vs. 60.9%; P = 0.036), protein (78.3 vs. 75.1%; P = 0.002), starch (98.9 vs. 99.2%. P = 0.026) as well as the total dietary fibre digestibility (39.8 vs. 35.7%; P = 0.011) as expected according to most data reviewed by Gidenne et al. (2012) and Birolo et al. (2016).
Table 1: Effect of cellobiose (CEL) supplementation and feeding plan (Ad libitum vs. Restricted - Rest.-) on apparent faecal digestibility at two different periods (39-43 d and 53-56 d of age) in growing rabbits.

<table>
<thead>
<tr>
<th>Feeding plan</th>
<th>CEL –</th>
<th>CEL+</th>
<th>SEM1</th>
<th>P-value2</th>
</tr>
</thead>
<tbody>
<tr>
<td>n1</td>
<td>Ad libitum</td>
<td>Rest.</td>
<td>Ad libitum</td>
<td>Rest.</td>
</tr>
<tr>
<td>39-43 d of age (restriction period)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Body weight, g</td>
<td>940</td>
<td>895</td>
<td>1121</td>
<td>937</td>
</tr>
<tr>
<td>DM intake, g/d</td>
<td>104</td>
<td>85.7</td>
<td>102</td>
<td>83.7</td>
</tr>
<tr>
<td>Faecal digestibility, %</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gross energy</td>
<td>60.9</td>
<td>62.8</td>
<td>60.8</td>
<td>64.7</td>
</tr>
<tr>
<td>Crude protein</td>
<td>75.5</td>
<td>77.2</td>
<td>74.6</td>
<td>79.4</td>
</tr>
<tr>
<td>Total dietary fibre5</td>
<td>35.8</td>
<td>38.3</td>
<td>35.6</td>
<td>41.3</td>
</tr>
<tr>
<td>Starch</td>
<td>98.8</td>
<td>99.1</td>
<td>99.0</td>
<td>99.2</td>
</tr>
<tr>
<td>53-56 d of age</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Body weight, g</td>
<td>2037</td>
<td>1949</td>
<td>2087</td>
<td>1904</td>
</tr>
<tr>
<td>DM intake, g/d</td>
<td>116</td>
<td>132</td>
<td>112</td>
<td>131</td>
</tr>
<tr>
<td>Faecal digestibility, %</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gross energy</td>
<td>63.3</td>
<td>62.4</td>
<td>61.8</td>
<td>62.5</td>
</tr>
<tr>
<td>Crude protein</td>
<td>73.9</td>
<td>73.7</td>
<td>72.4</td>
<td>74.7</td>
</tr>
<tr>
<td>Total dietary fibre5</td>
<td>41.9</td>
<td>39.3</td>
<td>38.2</td>
<td>38.9</td>
</tr>
<tr>
<td>Starch</td>
<td>99.1</td>
<td>99.2</td>
<td>99.3</td>
<td>99.0</td>
</tr>
</tbody>
</table>

Due to variance heterogeneity, a SEM value was included for each mean of the ad libitum groups, and another one for the means of restricted groups. The interactions Cellobiose × Feeding plan were not significant (P > 0.10). Number of rabbits used per treatment for digestibility from 39 to 43 d, and from 53 to 56 d of age, respectively. All values corrected for ash and protein.

Table 2: Effect of cellobiose supplementation (CEL) and feeding plan (Ad libitum vs. Restricted - Rest.-) on growth performance from 32 to 60 d of age of rabbits.

<table>
<thead>
<tr>
<th>Feeding plan</th>
<th>CEL –</th>
<th>CEL+</th>
<th>SEM1</th>
<th>P-value2</th>
</tr>
</thead>
<tbody>
<tr>
<td>n1</td>
<td>Ad libitum</td>
<td>Rest.</td>
<td>Ad libitum</td>
<td>Rest.</td>
</tr>
<tr>
<td>32-47 d of age (restriction period)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Body weight 32 d, g</td>
<td>726</td>
<td>683</td>
<td>713</td>
<td>712</td>
</tr>
<tr>
<td>Growth rate, g/d</td>
<td>61.3</td>
<td>49.5</td>
<td>62.3</td>
<td>48.9</td>
</tr>
<tr>
<td>Feed intake, g/d</td>
<td>127</td>
<td>90.8</td>
<td>124</td>
<td>89.2</td>
</tr>
<tr>
<td>Feed efficiency, g/g</td>
<td>0.488</td>
<td>0.546</td>
<td>0.504</td>
<td>0.548</td>
</tr>
<tr>
<td>Mortality</td>
<td>8.93</td>
<td>7.14</td>
<td>11.3</td>
<td>8.06</td>
</tr>
<tr>
<td>47-60 d of age</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Body weight 51 d, g</td>
<td>1627</td>
<td>1451</td>
<td>1642</td>
<td>1441</td>
</tr>
<tr>
<td>Growth rate, g/d</td>
<td>48.3</td>
<td>59.7</td>
<td>52.2</td>
<td>61.0</td>
</tr>
<tr>
<td>Feed intake, g/d</td>
<td>162</td>
<td>173</td>
<td>163</td>
<td>171</td>
</tr>
<tr>
<td>Feed efficiency, g/g</td>
<td>0.299</td>
<td>0.345</td>
<td>0.320</td>
<td>0.353</td>
</tr>
<tr>
<td>Mortality</td>
<td>7.14</td>
<td>1.79</td>
<td>9.68</td>
<td>3.23</td>
</tr>
<tr>
<td>32-60 d of age</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Body weight 59 d</td>
<td>2254</td>
<td>2230</td>
<td>2320</td>
<td>2233</td>
</tr>
<tr>
<td>Growth rate, g/d</td>
<td>55.2</td>
<td>54.4</td>
<td>57.6</td>
<td>54.5</td>
</tr>
<tr>
<td>Feed intake, g/d</td>
<td>143</td>
<td>129</td>
<td>142</td>
<td>127</td>
</tr>
<tr>
<td>Feed efficiency, g/g</td>
<td>0.388</td>
<td>0.421</td>
<td>0.406</td>
<td>0.427</td>
</tr>
<tr>
<td>Mortality</td>
<td>16.1</td>
<td>8.93</td>
<td>21.0</td>
<td>11.3</td>
</tr>
</tbody>
</table>

Due to variance heterogeneity, a SEM value was included for each mean of the Ad libitum groups, and another one for the means of Restricted groups. No interaction was found between Cellobiose × Feeding plan (P ≥ 0.133). Number of rabbits at the end of the fattening period and used to calculate growth traits. For mortality values the initial number of rabbits was 56, 56, 62, and 62, respectively. Morbid rabbits were not considered in the growth traits data: 3, 1, 7, and 4, respectively.

It might seem to be accounted for an increase in the mean retention time of the digesta (Gidenne et al., 2012), although it might also depend on the type of diet. During the restriction period feed intake of restricted rabbits was a 72% of that of the ad libitum group, while in the whole experimental period
accounted for a 90% of the *ad libitum* group. As expected, it decreased growth rate (56.4 vs. 54.5 g/d; \( P = 0.015 \)) and improved feed efficiency (0.424 vs. 0.397; \( P < 0.001 \)) in the whole period, reaching a lower final liveweight (\( P = 0.015 \)) but optimal for the market. Feed restriction, tended to reduce mortality rate (18.6 vs. 10.1%; \( P = 0.067 \)), and curiously no differences in mortality were observed during the restriction period, and this trend was due to the mortality reduction during the refeeding period (\( P = 0.041 \)). This result is similar to that reported by Farias-Kovac (2021), although this author reported a reduction in the mortality rate during the restriction period, and larger differences in the final liveweight between the *ad libitum* and restricted groups. It might be probably due to the shorter duration of the restriction period (-4 d) in this study. These results are mostly in agreement with those associated with feed restriction reviewed by Gidenne et al. (2012) and reported by Birolo et al. (2016).

There was no relevant interaction between CEL and feed restriction.

**CONCLUSIONS**

Cellbiose supplementation improved growth traits but had no influence on mortality. Gradual feed restriction, tended to decrease mortality with a minor influence of growth traits. No synergism was found was found between feed restriction and cellbiose supplementation.

**ACKNOWLEDGEMENTS**

This research was financed by the project MINECO-FEDER (AGL2015-66485-R and the pre-doctoral contract BES-2016-076649 obtained by C. Farias), and Comunidad de Madrid (technician contract PEJ-2017-TL/BIO-6777 obtained by Carla Izquierdo).

**REFERENCES**


Effect of cellobiose supplementation in drinking water and feed restriction on apparent faecal digestibility and growth performance in rabbits

Carlos E. Farías Kovac

12th World Rabbit Science Congress.
3-5 November, 2021.
Nantes, France
VFA production
Shift to beneficial bacteria
Direct effect on mucosa

Vegetal biomass

Refined oligosaccharides
* Enzymatically and/or chemically

Selective fermentation by intestinal microbiota
* Inclusion in the diet

Among 7 principal NDO’s used as prebiotics

- Cello-oligosaccharides (COS)
- Xilo-oligosaccharides (XOS)

- Some considered dietary fibre, different from NSP (Englyst et al., 2002)
- Selective effect on beneficial microbiota (Bielecka et al., 2002)

Some considered dietary fibre, different from NSP (Englyst et al., 2002)
Selective effect on beneficial microbiota (Bielecka et al., 2002)
**Cellobiose use**

Cello-oligosaccharides

Derived from cellulose

2 glucose molecules (β-1,4)

**Cellobiosidase**

**Cellulase**

7.0 g/L and 7.5 g/L x Low SF reduced mortality  
(Ocasio-Vega et al., 2018\(^a\), 2019)
Feed Restriction

Mortality
Feed efficiency
Growth performance

Non-consistent results or lack of effects

Progressive feed restriction??

(Gidenne et al., 2012)
(Gidenne et al., 2009; Romero et al., 2010)
(Crespo et al., 2020)
(Duperray and Guyonvarch, 2009; Birolo et al., 2016)
Objective

To evaluate the possible synergic effect of cellobiose supplementation with feed restriction on faecal digestibility and growth traits.
**MATERIAL Y METHODS**

**Factorial 2 x 2**

- Weaning: 32 days of age (BW = 700 ± 116 g)
- Diet: 21.1% CP; 35.5% NDF; 9.1% SF; 13.4% Starch

**Progressive feed restriction period**

**2 Cellobiose in drinking water (g/L)**

- 0.0 (Ocasio-Vega, 2018)
- 7.5

**Growth performance (32-60)**

- Carcass yield 10 / TTO
- Digestive organs.

**Faecal apparent digestibility (F.A.D)**
Progressive feed restriction program

Cellobiose supplementation along the whole fattening period

Animals restricted with a feed allotment of 50% respecting to control group

Restricted animals reached a feed allotment of 100% respecting to control group

Ad libitum feeding 9 days

(Duperray and Guyonvarch, 2009; Birolo et al., 2016)
No effect of Cellobiose was observed on mortality

- Why the lack of positive effect?
- Cellobiose intake in drinking water was equivalent to an inclusion in the diet of 1.5%.
- Levels of 2.2% increased mortality (Ocasio-Vega et al., 2018b)
- Cellobiose did not influence average feed intake.

![Average daily gain 47-60, g](image)

- **Cellulose** - **Cellulose** +
  - Average daily gain 47-60, g
  - **P**<sub>CEL</sub> = 0.0057
  - Cellobiose = 5%

![Feed efficiency 32-60d, g/g](image)

- **Cellulose** - **Cellulose** +
  - Feed efficiency 32-60d, g/g
  - **P**<sub>CEL</sub> = 0.003
  - Cellobiose = 3%
RESULTS

ACCUMULATED MORTALITY, %

- Restricted animals with a feed allotment of 100% respect to the ad libitum group
- Restricted animals with a feed allotment of 50% respect to the ad libitum group

CELL - / Ad libitum

CELL - / Restricted

CELL + / Ad libitum

CELL + / Restricted

AGE

AD. LIB.

RESTRICTED

Mortality 47-60d, %

- Ad libitum period

- Mortality 47-60d, % $P_{RA} < 0.041$

Mortality 32-60d

- Mortality 32-60d $P_{RA} < 0.067$

8% Ad libitum

3% Restricted

$P_{RA}$
**RESULTS**

**ADFI 32-60, g/d**

- **Restriction = -90%**
- **P<sub>RA</sub> = 0.001**
- AD. LIB.: 142.5 g/d
- RESTRICTED: 128.0 g/d

**ADG 32-60, g/d**

- **Restriction = -3%**
- **P<sub>RA</sub> = 0.001**
- AD. LIB.: 60.5 g/d
- RESTRICTED: 54.5 g/d

**Feed efficiency 32-60d, g/g**

- **Restriction = 7%**
- **P<sub>RA</sub> = 0.001**
- AD. LIB.: 0.397 g/g
- RESTRICTED: 0.424 g/g

**Final body weight, 59 d**

- **Restriction = -55, 98% A.L**
- **P<sub>RA</sub> = 0.015**
- AD. LIB.: 2286 kg
- RESTRICTED: 2232 kg
**CONCLUSIONS**

- **Cellobiose**: N.S. effect on Mortality
- **Feed restriction**: ↓ Mortality, ↑ Feed efficiency
- **Interaction**: No effect
Thank you.

Carlos E. Farias Kovac
CarlosEduardo_Farias@cargill.com

This research was financed by the project MINECO-FEDER (AGL2015-66485-R and the pre-doctoral contract BES-2016-076649 obtained by C. Farias), and Comunidad de Madrid (technician contract PEJ-2017-TL/BIO-6777 obtained by Carla Izquierdo).