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PINHEIRO V., GIDENNE T.

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DIGESTION AND HEALTH.**

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SUBSTITUTION OF WHEAT BY POTATO STARCH FOR GROWING RABBITS : EFFECT ON PERFORMANCES, DIGESTION AND HEALTH.

PINHEIRO V., GIDENNE T.*

UTAD, Secção de Zootecnia, apt. 201, 5000 Vila Real, Portugal

*Station de Recherches Cunicoles, INRA Toulouse, BP 27, 31326 Castanet- Tolosan, France.

ABSTRACT

Digestion and health status of the rabbit were measured according to a dietary addition of a resistant potato starch (PS) in replacement to wheat. The three experimental diets contained 0, 7 or 14% of PS (resp. WS-PS7-PS14), but had similar concentrations of starch (20%), proteins and fibre. Within 4 hours, 30% of the wheat starch was in-vitro hydrolysed, compared to 5% for PS. As PS level increased, the intake decreased by 11% (WS vs PS14) between 28-42 days of age. From weaning (28d) to slaughter (70d), weight gain was similar among diets (n=48/diet), while feed intake and feed conversion ratio increased with the PS incorporation. Mortality and morbidity rates were not significantly different between diets. However, the sanitary risk index (mortality+morbidity) tended to increase (P=0.107) with PS addition, particularly between 42 and 70d. of age (P= 0.073 for the contrast "WS vs PS7+PS14"). Starch faecal digestibility was 0.8 unit higher (P=0.04) for WS than for PS14 diet. In 6 weeks old rabbits, the ileal starch level increased (P<0.01) with PS addition : 0.3, 1.0, 1.4% resp. for WS, PS7 and PS14. The ileal starch level decreased with age (6 vs 10 wk.) only for diets PS7 and PS14 (P<0.05).

INTRODUCTION

A recent review on the digestion of starch in the rabbit (BLAS and GIDENNE, 1998) indicated that few studies have addressed the problem of its digestion in the small intestine, and particularly during the growth of the rabbit. Similarly, almost no results are available about the impact of the nature of starch on the non-specific enteritis of the growing rabbit. Consequently, the respective role of fibre and starch in the determinism of digestive troubles remain incompletely cleared.

The present study aimed to test if an increased quantity of starch (without an effect of the fibre level) entering the caecum could affect digestion and health status of the growing rabbit. A set of experiments was designed to measure several parameters of digestion and growth in 6 and 10 weeks old rabbits, fed iso-fibre diets containing either wheat (easily digestible) or purified crude potato starch (PS). The latter is acknowledged resistant to digestion in the small intestine of the monogastric animal, such as pig (MARTIN *et al.*, 1998).

MATERIALS AND METHODS.

Diets and experimental design

Three experimental diets were formulated (table 1), to obtain a linear replacement of wheat by purified crude potato starch "PS" (Roquette SA, Lestrem, France) : 0, 7, and 14%, respectively for diets WS, PS7 and PS14. Among diets, the level of starch, fibre and crude protein remain constant. The dietary ADF level of 16% corresponded to intermediate levels of fibre currently recommended (DE BLAS and MATEOS, 1998; GIDENNE, 2000). The level of starch was high (20%) to favour the potential effect of the starch nature. The ratio digestible crude protein/digestible energy was controlled to obtain a similar digestible protein supply among diets. Diets were given ad libitum in pelleted form (3.5mm diam.) from weaning (28 days of age) to slaughter (70d.), to NZW x Californian rabbits.

Total tract digestibility, health status, growth and starch level in the ileum.

Total tract apparent digestibility (TTAD) was measured according to E.G.R.A.N. (1995), on three groups of 10 rabbits, housed individually in metabolism cages from weaning (28d.) to

slaughter, and fed one of the experimental diets. TTAD was measured at 6 and 10 weeks of age (resp. between 37-41d., and 65-69days of age). Three further groups of 38 rabbits were used for measurements of growth performances. Health status was individually recorded three times a week from weaning to slaughter: all clinical signs of sickness (transitory diarrhoea, presence of mucus in excreta, abnormal behaviour, etc.) were registered. In addition, feed intake and live weight were measured weekly to detect any growth disturbances. Morbidity was defined as animals having abnormally low intake or growth ($< \text{mean} - 2.2 \text{ sd}$) or clinical signs of sickness, and they were not included in growth performances analyses. The sanitary risk index (SRI) was calculated as the sum of mortality and morbidity. The starch level in the terminal ileum (segment of 20cm prior to the ileo-caecal junction) was measured on samples of digesta were obtained from rabbits slaughtered at 42 and 70d old (10 samples per diet and per age).

Analytical methods

Organic matter (OM) was determined by ashing samples at 550°C for 5h. Measurements of fibre fractions (neutral detergent fibre: NDF, acid detergent fibre: ADF, acid detergent lignin: ADL) were made according to the sequential Van Soest procedure (Van Soest *et al.*, 1991) using an amylolytic pre-treatment with a thermostable amylase (AFNOR, 1997). Nitrogen was determined by DUMAS combustion method, and converted to crude protein (CP) using the factor 6.25. Starch was hydrolysed enzymically and the resultant glucose was measured by using the hexokinase G6PD-NAD system (Boehringer Mannheim). In vitro assay of wheat and potato starch

| Table 1: Composition of the experimental diets | | | | |
|---|----------------|-----------|------------|-------------|
| | Diets : | WS | PS7 | PS14 |
| <i>Ingredients (%)</i> | | | | |
| Potato Starch | | - | 7.0 | 14.0 |
| Wheat | | 31.0 | 20.5 | 10.0 |
| Soya | | 16.0 | 18.0 | 19.5 |
| Wheat bran | | 9.5 | 10.5 | 13.0 |
| Dehydrated lucerne meal | | 17.0 | 17.5 | 18.0 |
| Dehydrated beet pulp | | 19.0 | 19.0 | 18.0 |
| Wheat straw | | 5.5 | 5.5 | 5.5 |
| Minerals and Vitamins | | 2.0 | 2.0 | 2.0 |
| <i>Chemical analysis (g.kg air dry basis)</i> | | | | |
| Organic matter | | 853 | 845 | 846 |
| Crude protein (Nx625) | | 169 | 167 | 165 |
| Starch | | 198 | 195 | 200 |
| Neutral-detergent fibre (NDF) | | 327 | 320 | 305 |
| Acid-detergent fibre (ADF) | | 155 | 156 | 149 |

hydrolysis (figure 1) were performed at 37°C, according to TOLLIER and GUILBOT (1974) and SALOMONSSON *et al.* (1984), by measuring the resultant reducing sugars according to the DNS method (MILLER, 1959).

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Statistical analysis

Data on growth and feed intake were examined by one-way analysis of variance using the GLM procedure of SAS (SAS, 1988). Catmod procedure (SAS) was utilised for data of morbidity and mortality. Results of TTAD and ileal starch were subjected to a two factorial (age and diet) analysis of variance, and means were compared using the Scheffe test.

RESULTS

As expected, feeds contained similar concentrations of starch, proteins and fibre (table 1). The in-vitro hydrolysis of the two types of starch revealed large differences (figure 1): within 4 hours wheat starch was hydrolysed at 30%, compared to less than 5% for PS.

Feed intake, growth, and health status

With the increase of the potato starch level, the voluntary intake of animals was significantly increase by 11% during the two weeks postweaning (table 2) and by 8% during the finishing

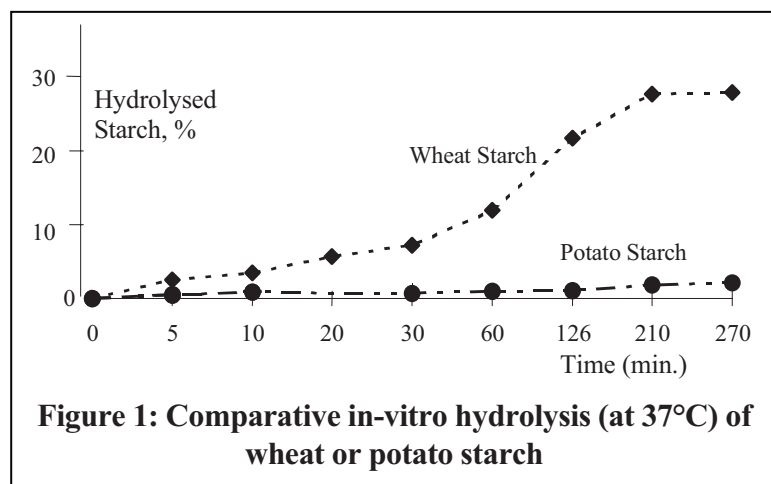


Figure 1: Comparative in-vitro hydrolysis (at 37°C) of wheat or potato starch

fattening period (42-70d. old). The weight gain increased linearly with the incorporation of PS (+14% for PS14, $p < 0.05$), during the postweaning period. Between 42 and 70 d. old, the feed conversion ratio was 9% higher for PS14 group, mainly because of the increase of the feed intake since weight gain remain unaffected. Over the complete growth period, weight gain was similar among diets, while feed intake and feed

conversion ratio increased with the PS incorporation.

Mortality and morbidity rates were not significantly different between diets. However, while the mortality rate (whole period) remained relatively low for WS, it tended to increase with PS diets ($P = 0.15$ for the contrast "WS vs PS7+PS14", table 3). Similarly, SRi tended to increase with PS addition particularly between 42 and 70d. old ($P = 0.0703$, for the contrast "WS vs PS7+PS14").

Table 2: Feed intake and growth performances of rabbits from weaning (28d) to slaughter (70d), according to type of starch (n=48/diet).

| Diets | WS | PS7 | PS14 | SEM ¹ | P level | EfLin ² |
|------------------------------|--------|---------|--------|------------------|---------|--------------------|
| Live weight at weaning (g) | 566 | 566 | 568 | 13 | ns | |
| Live weight at 42 d (g) | 1123 | 1159 | 1202 | 26 | 0.09 | 0.03 |
| Live weight at slaughter (g) | 2221 | 2298 | 2294 | 42 | ns | |
| <i>Weaning - 42d old</i> | | | | | | |
| Daily feed intake (g) | 63.8 b | 65.0 b | 70.5 a | 1.9 | 0.03 | <0.01 |
| Daily weight gain (g) | 39.4 b | 42.0 ab | 44.9 a | 1.2 | 0.07 | <0.01 |
| Feed conversion ratio | 1.60 | 1.56 | 1.59 | 0.02 | ns | |
| <i>42d - 70d old</i> | | | | | | |
| Daily feed intake (g) | 106 | 111 | 114 | 5.6 | 0.09 | 0.03 |
| Daily weight gain (g) | 39.4 | 40.0 | 38.8 | 0.9 | ns | |
| Feed conversion ratio | 2.72 b | 2.77 ab | 2.95 a | 0.05 | <0.01 | <0.01 |
| <i>Weaning at slaughter</i> | | | | | | |
| Daily feed intake (g) | 92.1 b | 95.9 ab | 99.9 a | 2.1 | 0.03 | <0.01 |
| Daily weight gain (g) | 39.5 | 41.0 | 41.0 | 0.8 | ns | |
| Feed conversion ratio | 2.31 b | 2.34 ab | 2.43 a | 0.03 | 0.01 | <0.01 |

1 - SEM: stand error of the mean ; ns = $P > 0.15$; ^{a,b} - Means having a common letter did not differ significantly at level $P = 0.05$; 2 - EfLin: linear effect

Digestion in the whole tract and starch concentration in the ileum of growing rabbits.

Except for starch, total tract apparent digestibility (TTAD) was not greatly affected by the level of PS, although we detected for OM a trend ($P = 0.09$) for a slight linear reduction (table 4). In return, we recorded a 0.8 unit reduction for starch digestibility ($P = 0.04$) between WS and PS14 group (linear effect : $P < 0.01$), while it did not differ in 6 or 10 wks old rabbits.

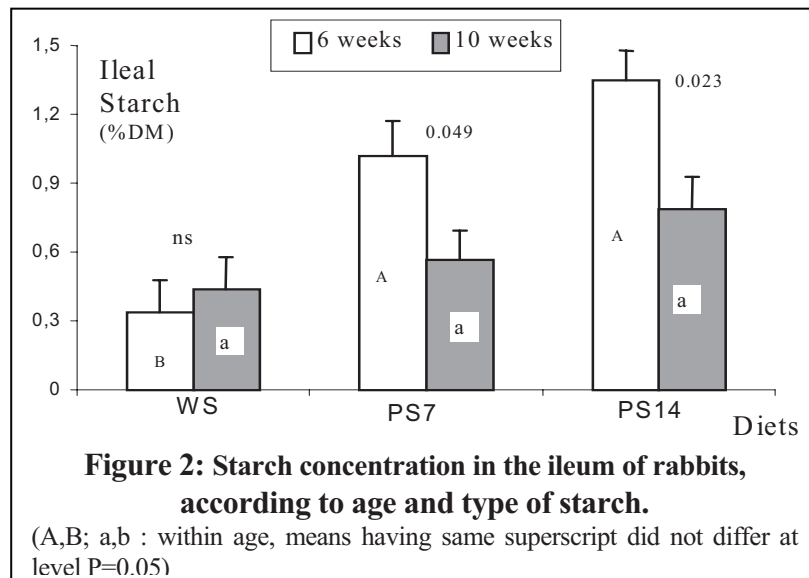
Crude protein digestibility was reduced by 6% ($P < 0.001$) between 6 and 10 weeks of age, while fibre (NDF, ADF) tended to be more digested in 10wks old rabbits.

A significant interaction ($P < 0.05$) between the effect of the age (6 vs 10 weeks old) and the effect of the diets was detected for the ileal starch concentration, therefore we present our results according to age and fibre level in figure 2. In 6 weeks old rabbits (2 wks after weaning), the starch level increased sharply ($P < 0.01$) from WS to PS7, but remained inferior to 1.5% even with 14% of PS in the diet. At 10 weeks of age, ileal starch remained similar among diet (mean 0.5%). The starch concentration in the ileum decreased with age (6 vs 10 wk.) in the diets PS7 and PS14 ($p < 0.05$).

DISCUSSION

Effect of age

Starch digestion in the whole tract appeared always very high (over 95%), even for young rabbits around weaning, as starch escaping the enzymatic digestion in the small intestine is rapidly fermented by the caecal flora (BLAS *et al.*, 1990 ; PARIGI BINI *et al.*, 1990 ; GIDENNE and PEREZ, 1993a ; MAERTENS and LUZI, 1995). Hence, the starch losses in faeces in young rabbits around weaning is inferior to 1% with feeds containing barley starch, but it could reached 6% with more resistant starch such as maize (GIDENNE and PEREZ, 1993a).



However, it is more relevant to examine starch digestion before caecal flora activity. Blas *et al.* (1994) found around 7% of starch in ileum of 5 weeks old rabbits. We here measured a lower level of ileal starch ($< 1.3\%$), but for rabbits one week older. For finishing growing rabbits (9-10 wks old) PINHEIRO and GIDENNE (1999) estimated that the starch flow at the ileum remained lower than 2g/d even with high starch diet

(30%), due to low ileal starch concentration ($< 3\%$). However, the measurement of starch in ileal digesta of the growing rabbit remains difficult, as low quantity of sample are available. Moreover, animals are fed ad libitum, leading to various pattern of intake and thus favour a high interindividual variability ($\approx 20\%$). Therefore, unreliability among previous results is not surprising. We found about 0.5% ileal starch for finishing rabbits fed WS feed, while with a very similar diet PINHEIRO and GIDENNE (1999) reported a level of 1.4%. Conversely in adult rabbits, GIDENNE and PEREZ (1993) reported a 0.6% ileal starch level with high starch feeds (25%). The effect of age obviously interacts with the nature of the starch. Thus, we detected higher ileal losses of starch for 6 weeks than for 10 wks old rabbits only with feed containing potato starch. This was also in agreement with GIDENNE *et al.* (1999) indicating similar ileal level of starch for 6 or 10 wks old rabbits fed a diet containing wheat starch (less resistant).

Effect of starch nature

The potato starch is acknowledged as resistant to the digestion in the small intestine. For example, 56% of the same type of potato starch (Roquette SA) escaped digestion in the small intestine of the pig, fed a diet with only 6.5% of PS (MARTIN *et al.*, 1998). Our results of in-vitro hydrolysis of starch confirmed that this purified potato starch was highly resistant to

amylase compared to wheat starch. However, the ileal starch level in rabbits receiving PS was surprisingly low ($\approx 1.2\%$), suggesting that more than 90% of the starch was digested before caecum, even when the two third of starch intake corresponded to potato (diet PS14). This contrasted with in-vitro results, and even suggested that starch is digested in the intestine not only by endogenous amylase (SCAPINELLO *et al.*, 1999), but also by bacterial amylase provided by soft faeces, as previously observed in the stomach (HÖRNICKE and MACKIEWICZ, 1976). In spite of this high starch digestion in the intestine, overall digestion appeared lower when PS replaced wheat, and was associated with a higher feed intake and feed conversion ratio. The consequences of incorporating resistant starch on health status of the growing rabbits seemed weak. Nevertheless, with PS addition, slightly higher ileal starch level seemed associated to a tendency for a higher sanitary risk. The latter must be confirmed on larger group of rabbits. In conclusion, the impact of starch entering the caecum on health of the growing rabbit failed to be completely elicited, as ileal starch digestion remain very high even with a dietary addition of a resistant starch. Reversely, this high intestinal efficiency to digest starch supports the hypothesis that fibre supply, rather than starch overload, plays a key role in the determinism of non-specific enteritis.

Table 3: Health status of the growing rabbit, according to type of starch.

| | Diets | WS | PS7 | PS14 | <i>P level</i> | <i>Contrast *</i> |
|-----------------------------------|-------|------|------|------|----------------|-------------------|
| <i>Weaning (28d.) to 42d. old</i> | | | | | | |
| Mortality (%) | | 4.2 | 10.4 | 2.1 | <i>ns</i> | <i>ns</i> |
| Morbidity (%) | | 14.6 | 12.5 | 18.8 | <i>ns</i> | <i>ns</i> |
| SRi (%) | | 18.8 | 22.9 | 20.9 | <i>ns</i> | <i>ns</i> |
| <i>42 to 70d. old (slaughter)</i> | | | | | | |
| Mortality (%) | | 0.0 | 7.9 | 7.9 | <i>ns</i> | <i>ns</i> |
| Morbidity (%) | | 10.4 | 13.1 | 23.7 | <i>ns</i> | <i>ns</i> |
| SRi (%) | | 10.4 | 21.0 | 31.7 | <i>0.107</i> | <i>0.073</i> |
| <i>Weaning to slaughter</i> | | | | | | |
| Mortality (%) | | 4.2 | 18.3 | 10.0 | <i>0.14</i> | <i>0.15</i> |
| Morbidity (%) | | 25.0 | 25.6 | 42.5 | <i>ns</i> | <i>ns</i> |
| SRi (%) | | 29.2 | 43.9 | 52.5 | <i>ns</i> | <i>0.12</i> |

SRi = sanitary risk index = mortality + morbidity rate; initial number of animal per diet = 48 ; ns = $P > 0.15$
 *: diet WS vs (PS7+PS14)

Table 4: Total tract apparent digestibility (TTAD) of major nutrients, according to age and type of starch.

| TTAD (%) | Diets | | | Age (weeks) | | <i>SEM</i> ¹ | <i>P level</i> | | |
|----------------|-------------------|--------------------|-------------------|-------------|------|-------------------------|----------------|-----------------|------------|
| | WS | PS7 | PS14 | 6 | 10 | | Diet | Age | <i>D*A</i> |
| Organic matter | 72.8 | 72.2 | 71.0 | 72.0 | 71.9 | <i>0.7</i> | <i>ns</i> | <i>ns</i> | <i>ns</i> |
| Crude protein | 79.6 | 80.3 | 77.9 | 81.8 | 76.6 | <i>0.6</i> | <i>0.051</i> | <i><0.01</i> | <i>ns</i> |
| NDF | 41.0 | 40.4 | 38.4 | 38.4 | 41.5 | <i>1.5</i> | <i>ns</i> | <i>0.11</i> | <i>ns</i> |
| ADF | 29.8 | 28.3 | 28.1 | 26.7 | 30.9 | <i>1.8</i> | <i>ns</i> | <i>0.08</i> | <i>ns</i> |
| Starch | 99.8 ^a | 99.5 ^{ab} | 99.0 ^b | 99.4 | 99.5 | <i>0.1</i> | <i>0.04</i> | <i>ns</i> | <i>ns</i> |

1, ns: see table 2

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