

## Digestibility indices and N balance in growing rabbits fed a basal diet of water spinach (*Ipomoea aquatica*) supplemented with broken rice

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### Abstract

Four Local and 4 New Zealand White male rabbits with an initial live weight of  $1.75 \pm 0.10$  kg were fed water spinach (*Ipomoea aquatica*) ad libitum supplemented with broken rice (0, 4, 8 and 12g/day) according to a duplicate 4\*4 Latin Square design.

There were no significant differences for the interaction genotype x diet in any of the indices examined. There was a significant decrease in faecal pH with increasing levels of broken rice. However, there was no significant effect among treatments on the DM, NH<sub>3</sub> and SCFA concentrations in the faeces. Faecal DM content was higher for local than for NZ White rabbits.

There were no significant differences in digestibility indices between levels of broken rice. There was a trend ( $P < 0.10$ ) for DM and organic matter digestibility indices to be higher in the local than in the NZ genotype. This trend was significant ( $P < 0.05$ ) in N digestibility favouring the local breed. There was no significant effect on N retention due to genotype or the level of broken rice.

It is concluded that fresh water spinach has a high nutritive value for rabbits. There were no nutritional advantages from providing additional digestible energy in the form of broken rice. Water spinach appears to be a complete diet for rabbits providing water as well as all required nutrients.

**Key words:** Broken rice, digestibility, N balance, rabbits, water spinach (*Ipomoea aquatica*).

### 1. Introduction

Water spinach (*Ipomoea aquatica*) appears to have a high potential as a feed source for rabbits according to the preliminary report of Hongthong Phimmasan *et al.* (2004). It is a vegetable that is consumed by people and animals. It has a short growth period, is resistant to common insect pests and can be cultivated either in dry or flooded soils. Moreover, it has been found that water spinach is a vegetable with a high potential to efficiently convert nitrogen from biogas digester effluent into edible biomass with high protein content (Kean Sophea and Preston, 2001).

Water spinach was reported to have a high *in vitro* N digestibility when incubated in rumen fluid (68.8%) and a high concentration of water extractable N (52.0%) (Ly and Preston, 2001). In the preliminary trial conducted by Hongthong Phimmasan *et al.* (2004), using the indirect “insoluble ash” method to assess the digestibility of its components (Van Keulen and Young, 1977), it was

found that water spinach had high digestibility indices of DM, ash, organic matter and crude protein (84.7, 76.7, 88.4, and 79.6%, respectively) for growing rabbits.

Broken rice is commonly used as an energy supplement source for pigs and poultry in Cambodia, and can be found easily in local markets. Moreover, its use for this purpose does not compete with human food, due to a current surplus of rice grain production in the country.

The aim of this experiment was to confirm the observations of Hongthong Phimmasan *et al.* (2004), concerning the nutritive value of water spinach in growing rabbits, but using the direct method for determining digestibility. It was hypothesized that increasing levels of broken rice will improve the digestibility coefficients and N balance of rabbits.

## 2. Materials and Methods

### 2.1. Location and climate

The experiment was conducted in the ecological farm of the Center for Livestock and Agriculture Development (CelAgrid-UTA Cambodia), located in Rolous village, Rolous commune, Kandal Stoeung district, Kandal province, about 26 km from Phnom Penh City, Cambodia. During the trial (10 July to 19 August, 2004), the average ambient temperature was  $25.9 \pm 0.96$  °C in the morning at 6: 00 am,  $31.7 \pm 1.59$  °C in the middle of the day (12:00 am) and  $28.3 \pm 1.90$  °C in the afternoon at 6: 00 pm.

### 2.2. Experimental design

Four local male rabbits ( $1.77 \pm 0.10$  kg live weight) and four New Zealand White males ( $1.74 \pm 0.10$  kg) were allocated to 4 experimental treatments according to a duplicate 4\*4 Latin square arrangement (Table 1).

The treatments were:

- Br0: water spinach ad libitum
- Br4: same as Br0 but with 4 g/day of broken rice
- Br8: same as Br0 but with 8 g/day of broken rice
- Br12: same as Br0 but with 12 g/day of broken rice

**Table 1.** Layout of experiment.

Period/rabbit	Local				New Zealand			
	1	2	3	4	5	6	7	8
1	BR4	BR8	BR0	BR12	BR0	BR12	BR4	BR8
2	BR8	BR12	BR4	BR0	BR12	BR8	BR0	BR4
3	BR0	BR4	BR12	BR8	BR8	BR4	BR12	BR0
4	BR12	BR0	BR8	BR4	BR4	BR0	BR8	BR12

The rabbits were housed in metabolism cages during the trial. The metabolism cages (50 x 55cm) were built to allow the quantitative collection of hard faeces and urine (Photo 1), and were installed in an open floor. Each experimental period consisted of five preliminary days when the rabbits were adapted to the diets followed by another five days for collection of faeces, urine and

feed refusals. The rabbits were weighed at the beginning of the trial and at the end of each period.



**Photo 1.** Metabolism cages for rabbits designed to collect faeces and urine.

### 2.3. Feeds and feeding system

The water spinach was collected along the canal of CelAgrid-UTA Cambodia. The whole plant (combined stems and leaves) was offered as bunches hanging from the side of the cage (Photo 1). The proportions of stems and leaves were determined in samples taken at random on four occasions during the trial (Table 2). The level offered was approximately 50% greater than the recorded intake. Fresh water spinach was offered three times per day: in the morning at 7.30 am, at the middle of the day at 12:00 am and in the afternoon at 4:00 pm. On the basis of previous observations (Miech Phalla, personal communication), additional water was not provided as the fresh plant contains almost 90% water thus providing a ratio of moisture to dry matter of close to 9:1.

The broken rice was purchased in a local market, about 5 km from CelAgrid-UTA Cambodia. It was offered in a metal bowl anchored by wire to the side of the cage to avoid spillage. The broken rice was offered two times per day, in the morning at 7: 30 am and in the afternoon at 4:00 pm, in order to decrease the speed of eating by the rabbits.

**Table 2.** Proportion of leaves and stems contained in water spinach foliage (% in dry basis).

	<b>Proportion</b>
Leaves	46.1 ± 2.11 <sup>1</sup>
Stems	53.9 ± 2.11

<sup>1</sup> Mean and standard error of four determinations

#### 2.4. Data collection

Feed refusals and faeces were collected every day and were kept frozen in plastic bags until analysis. At the end of each period, feed refusals and faeces were mixed thoroughly by hand and a representative sample homogenized in a coffee grinder, for analysis of pH, NH<sub>3</sub>, Short Chain Fatty Acids (SCFA), DM, N, crude fiber and ash content. Organic matter concentration was calculated as 100 minus % ash in dry basis. Urine was collected in a plastic bucket to which 40% (w/v) sulphuric acid was added to maintain the pH below 4.0. At the end of each period the volume was measured and a sample analysed for N.

#### 2.5. Chemical analyses

Chemical analyses of diets and faeces were undertaken following the methods of AOAC (1990) for ash, N, and crude fiber. The DM content was determined using the microwave method of Undersander *et al.* (1993). Fresh faeces were analysed for pH by a digital meter with glass electrode. Faecal short chain fatty acids (SCFA) and ammonia were determined by titration of the sample recovered after steam distillation of filtered faecal slurry (1:5 by weight of fresh faeces and water) as outlined by Ly *et al.* (2001). The N content of urine was determined by the AOAC (1990) procedure.

#### 2.6. Statistical analyses

The data were subjected to analyses of variance according to the general linear model of the Minitab software (Minitab release 13.31, 2000). When the “F” test was significant ( $P < 0.05$ ), the means were separated using the Tukey comparison option in the Minitab software. The model used was the following:

$$Y_{hijk} = \mu + B_h + T_i + P_j + A_k + e_{hijk}$$

- $Y_{hijk}$  = Dependent variable
- $\mu$  = overall mean
- $B_h$  breed effect
- $T_i$  = treatment effect
- $P_j$  = period effect
- $A_k$  = animal effect
- $B_h T_i$  interaction of breed and treatment
- $e_{hijk}$  = random error

### 3. Results and discussion

#### 3.1. Animal status

No signs of discomfort were apparent during the conduct of the trial, and the animals showed a good health and gained in live weight.

### 3.2. Feed characteristics

There was a low content of ash, N and crude protein in the broken rice (Table 2). N and crude protein concentrations were higher in leaves than in stems of water spinach. Crude fiber content was higher in the stems than in leaves.

**Table 3.** Feed characteristics (% in DM, except for DM which is on fresh basis).

	DM	N	Crude protein	Ash	Organic matter	Crude fiber
Broken rice	88.6	1.02	6.37	1.19	98.8	-
<b>Water spinach</b>						
Leaves	11.6	5.62	35.1	10.9	89.1	8.58
Stems	6.87	3.28	20.5	17.8	82.2	17.2
Crude protein equals N*6.25						

### 3.3. Feed intake

The rabbits selected more leaves than stems (Figure 1). There were no significant differences among treatments in total DM intake (Table 4). However, DM intake from stems plus leaves and the intake of crude protein, declined as the level of broken rice increased ( $P<0.01$ ). The introduction of increasing levels of broken rice in the ration provoked a significant ( $P<0.01$ ) decrease in total crude protein intake by the animals.

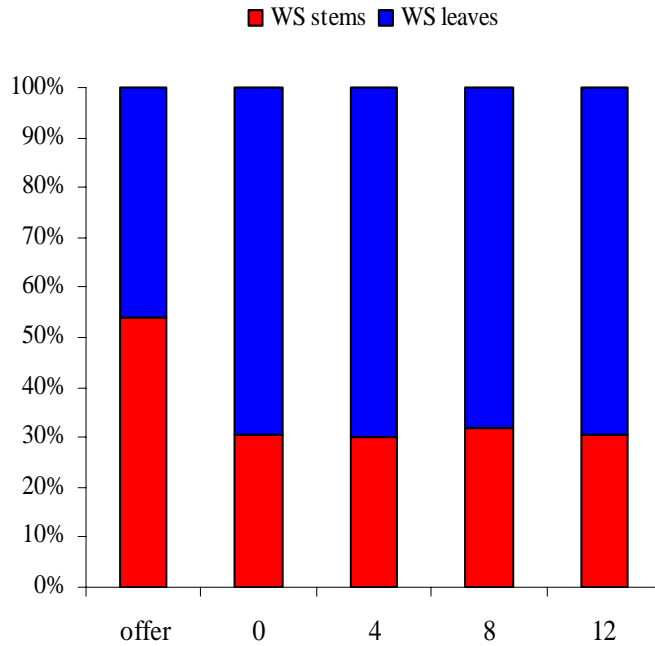
Rabbits of the local breed selected more leaves than stems and had higher intakes of DM and crude protein, compared with the NZ rabbits. DM intake per unit live weight was higher with 4 and 12 g broken rice compared with zero and 8 g/day ( $P<0.05$ ) and higher for the local compared with the NZ breed ( $P<0.01$ ). The range of intakes (40 to 42 g DM/kg live weight) are slightly lower than was reported by Hongthong Phimmasan *et al.* (2004) on similar diets (48 to 66 g DM/kg live weight).

**Table 4.** Mean values for feed intake (g/day) of rabbits fed water spinach ad libitum and graded levels of broken rice.

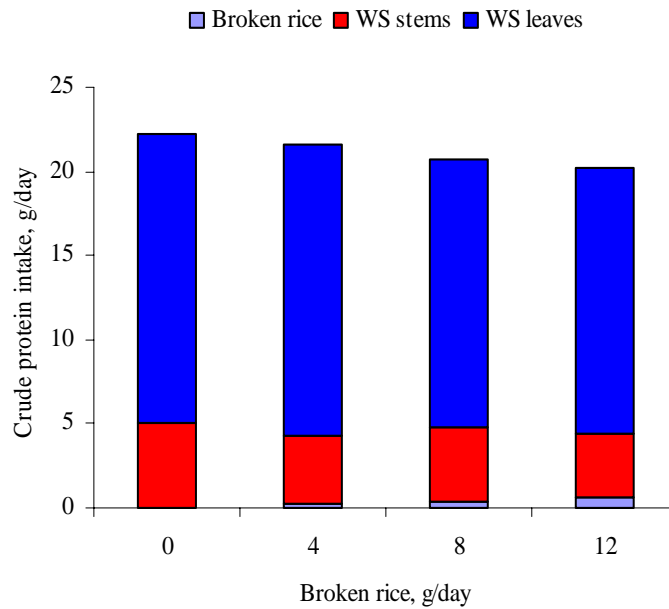
	0	Broken rice, g/day				SEM	LB	Genotype	
		4	8	12	NZWB			SEM	
<b>DM</b>									
Leaves	49.5	50.3	45.7	45.6	1.54 <sup>+</sup>	51.5	44.0	1.03 <sup>***</sup>	
Stems	21.8	21.7	21.3	20.1	1.09	19.6	22.9	0.75 <sup>**</sup>	
Leaves plus stems	71.3	72.0	67.0	65.7	1.79 <sup>**</sup>	71.1	66.9	1.27 <sup>**</sup>	
Broken rice	0.00	3.54	7.08	10.6	0.01 <sup>***</sup>	5.31	5.31	0.45	
Total	71.3	75.6	74.1	76.4	1.79	76.5	72.2	1.35 <sup>*</sup>	
As g/kg live weight	39.6	42.0	39.6	42.4	0.69 <sup>*</sup>	42.5	40.1	0.485 <sup>**</sup>	
<b>Crude protein</b>									
Leaves	17.1	17.3	16.0	15.7	0.431 <sup>*</sup>	17.8	15.2	0.308	
Stems	5.06	4.06	4.31	3.75	0.250 <sup>**</sup>	4.06	4.44	0.188 <sup>**</sup>	
Broken rice	0.00	0.25	0.44	0.69	0.063 <sup>***</sup>	0.31	0.31	0.063	
Total	22.4	21.9	21.0	20.4	0.375 <sup>**</sup>	22.6	20.3	0.250 <sup>**</sup>	

<sup>+</sup>  $P<0.10$ ; <sup>\*</sup>  $P<0.05$ ; <sup>\*\*</sup>  $P<0.01$ ; <sup>\*\*\*</sup>  $P<0.001$

LB: Local Breed and NZWB: New Zealand White Breed.



**Figure 1.** Proportions of leaves and stems (DM basis) in water spinach offered and consumed.



**Figure 2.** Quantities of crude protein consumed in leaves and stems of water spinach and in broken rice.

### 3. 4. Faecal characteristics and output

There was a significant ( $P < 0.05$ ) decrease in faecal pH when broken rice levels were increased (Table 5). This was related with a significant ( $R^2 = 0.999$ ) curvilinear increase in SCFA concentration (Figure 3). The increase of SCFA in the faecal output is in accordance with the

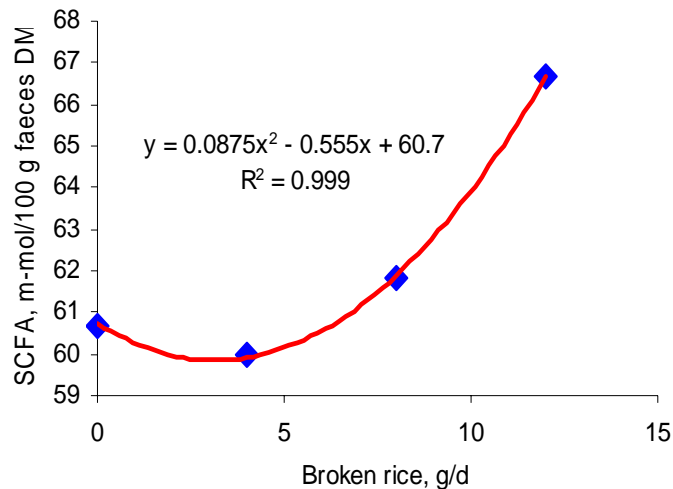
findings of Hongthong Phimmasan *et al.* (2004), who reported that the increase of SCFA was probably due to the fermentation in the caecum and large intestine of the starch in the broken rice. Bellier *et al.* (1995) described the relationship between pH and SCFA, with the caecal pH varying inversely to the increase in SCFA concentration. Gidenne and Perez (1993) showed that faecal losses of starch were very low in most cases, although as levels of starch increased in the diet, those losses were significantly greater. Starch digestion normally occurs in the small intestine; however, De Blas and Gidenne (1998) reported that it may also be degraded to some extent in other parts of the digestive tract such as the stomach and the large intestine.

**Table 5.** Faecal characteristics and excretion in rabbits fed water spinach ad libitum and graded level of broken rice.

	Broken rice, g/day				SEM	LB	Genotype	
	0	4	8	12			NZWB	SEM
<b>Faecal characteristics</b>								
Faecal pH	7.02	6.67	6.61	6.49	0.13*	6.49	6.60	0.093
DM, %	36.9	37.4	37.4	37.0	2.04	39.1	34.7	1.44*
SCFA, mmol/100 g DM	60.7	60.0	61.8	66.7	5.67	60.9	63.7	4.01
NH <sub>3</sub> , mmol/100g DM	21.0	20.8	24.8	24.9	2.86	21.2	24.4	2.02
<b>Faecal excretion, g/kg DM intake</b>								
DM	195	192	188	167	10.0	176	195	6.97 <sup>+</sup>
Fresh material	540	523	517	466	36.0	454	569	25.5 <sup>**</sup>
Water	346	331	329	299	30.0	278	375	21.4 <sup>**</sup>
<b>Faecal excretion, mmol/kg DM intake</b>								
SCFA	116	114	113	112	10.0	105	122	7.19
NH <sub>3</sub>	39.9	39.5	46.6	41.4	5.34	37.4	46.3	3.78

<sup>+</sup>  $P < 0.10$ ; \*  $P < 0.05$ ; \*\*  $P < 0.01$

LB: Local Breed and NZWB: New Zealand White Breed.



**Figure 3.** Relationship between intake of supplementary broken rice and concentration of short chain fatty acids in the faeces of rabbits fed ad libitum water spinach.

### 3. 5. Digestibility indices and N balance

There were no significant differences in digestibility indices due to feeding of supplementary broken rice. However, there was a tendency for digestibility of DM ( $P=0.066$ ) and organic matter ( $P=0.091$ ) to be higher in the Local breed as compared to the New Zealand genotype. The difference was significant for crude protein ( $P=0.028$ ) in favour of the local breed. These differences between the breeds can be explained by the greater selection for leaves, compared with stems, by the local breed (Table 3), and the fact that digestibility of DM and especially of crude protein were related to the proportion of leaves consumed (Figures 4 and 5). There were no differences in digestibility of crude fiber between the two breeds; however, the fiber intake by the local breed would be less because there is much less fiber in the leaves than in the stems (Table 3).

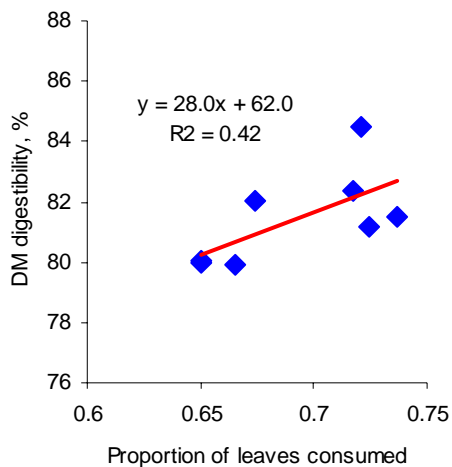
The digestibility coefficients in the present study were in the same range as those reported by Hongthong Phimmasan *et al.* (2004) (84 to 89% for DM and 76 to 80% for crude protein). They are higher than has been observed with commercial pelleted diets fed to local rabbits in Mauritius (69.2% for DM and 74% for crude protein) (Ramchun *et al.*, 2000a), or a combination of Star grass with mash (68.8% for DM and 81.3% for crude protein) (Ramchun *et al.*, 2000b). Bamikole and Ezenwa (1999) in Nigeria reported 50.2% for DM and 61.4% for crude protein for a diet of Verano stylo and 52.8% for DM and 61.8% for crude protein with Guinea grass. Cunha *et al.* (2004) in Portugal offered alfalfa as a source of fiber and recorded 61.4% digestibility for DM and 73.2% for crude protein.

**Table 6.** Digestibility indices and N balance in rabbits fed water spinach ad libitum and graded level of broken rice.

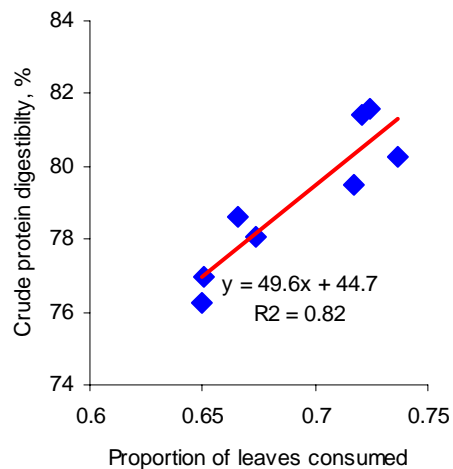
	Broken rice, g/day					Genotype		
	0	4	8	12	SEM	LB	NZWB	SEM
<b>Digestibility, %</b>								
DM	80.5	80.8	81.2	83.3	1.03	82.4	80.5	0.72 <sup>+</sup>
Crude protein	80.1	78.6	77.9	79.8	1.26	80.7	77.5	0.89 <sup>*</sup>
Ash	72.6	73.2	71.2	75.0	1.82	73.7	72.3	1.28
Organic matter	80.8	80.7	81.5	83.5	1.01	82.5	80.8	0.71 <sup>+</sup>
Crude fiber	64.8	63.1	62.9	64.4	2.43	63.6	64.0	1.67
<b>N retention</b>								
In g/day	1.09	1.00	0.98	1.02	0.18	1.13	0.91	0.12
As % of intake	31.1	29.8	30.5	32.3	4.56	32.9	29.0	3.08
As % of digestion	38.7	38.4	39.8	40.8	5.18	40.9	37.9	3.52

<sup>+</sup>  $P<0.10$ ; <sup>\*</sup>  $P<0.05$

LB: Local breed; NZWB: New Zealand White breed



**Figure 4.** Relationship between proportion of leaves of water spinach consumed and DM digestibility.



**Figure 5.** Relationship between proportion of leaves of water spinach consumed and crude protein digestibility.

There were no differences in any of the measurements of N retention due to broken rice supplementation and no differences between breeds (Table 6).

#### 4. Conclusions

- Fresh water spinach has a high nutritive value for rabbits as shown by the high voluntary intake, and high apparent digestibility of DM and crude protein.
- There were no nutritional advantages from providing additional digestible energy in the form of broken rice
- With an offer level of about 50% over recorded intakes, the rabbits selected strongly for leaves rather than stems.
- Water spinach appears to be a complete diet for rabbits providing water as well as all required nutrients.

#### 5. Acknowledgements

The authors would like to thank the Swedish Agency for Research Cooperation with Developing Countries (SAREC) for funding this study through the regional MEKARN project. In addition, thanks are given to Mr. Von Vyreak and Miss Chuon Vasna for their assistance during the conduct of the experiment. We address our gratitude to Mr. Chhay Ty for help with analyses in the laboratory.

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