

# Growth and survival of reared Cambodian field crickets (*Teleogryllus testaceus*) fed weeds, agricultural and food industry by-products

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## Abstract

This study evaluated survival and growth of Cambodian field crickets (*Teleogryllus testaceus*) during captivity when fed a set of local weed species, agricultural and food industry by-products. Wild individuals were caught at two locations in Cambodia, kept in pens and fed commercial chicken feed until the second generation off-spring hatched. First larval stage nymphs from this generation were collected and used in a 70-day feeding trial with one control treatment (chicken feed) and 12 experimental treatments (rice bran, cassava plant tops, water spinach, spent grain, residue from mungbean sprout production, and *Alternanthera sessilis*, *Amaranthus spinosus*, *Commelina benghalensis*, *Cleome rutidosperma*, *Cleome viscosa*, *Boerhavia diffusa* and *Synedrella nodiflora*). The crickets were kept in plastic cages and feed intake, weight and survival of crickets were recorded weekly. Overall survival did not differ between chicken feed and the experimental treatments with the exception of crickets fed *B. diffusa*, which had lower survival. From day 35 to day 49, survival on *A. sessilis* was also lower ( $P < 0.05$ ) than on chicken feed. There was no difference in weight between crickets fed chicken feed, cassava tops and *C. rutidosperma*. However, crickets fed *A. sessilis*, *A. spinosus* and *B. diffusa* weighed less than those fed chicken feed already at day 21. The feed conversion rate ranged from 1.6 to 3.9 and was  $\leq 1.9$  in crickets fed chicken feed, cassava plant tops and *C. rutidosperma*. Thus this study shows that it is possible, using simple means, to rear Cambodian field crickets. Cassava plant tops and *C. rutidosperma* both have great potential as cricket feed and the other weeds, with the exception of *A. sessilis*, *A. spinosus* and *B. diffusa*, agricultural and food industry by-products tested, also showed potential.

**Keywords:** entomophagy, cassava, *Cleome rutidosperma*, feed conversion, weight

## 1. Introduction

Poverty and malnutrition are major interlinked problems in Cambodia (<http://tinyurl.com/arr9k5>). To alleviate the situation, there is a need for production of nutritious food products. In this context, entomophagy could be one option to improve nutrition, by providing high-quality animal protein. Crickets are popular as food in Cambodia and the market has been described as fast growing (Münke, 2012). The main supply of crickets to the market is currently from light trap owners harvesting wild crickets (Münke, 2012). However, this practice is not likely to meet future

year-around demand for crickets. Moreover, catching wild crickets could be a potential threat to existing wild cricket populations. Therefore, sustainable cricket rearing systems that preferably require limited investment and have low feed costs should be developed. Sustainability should permeate both the areas of feed resources and choice of species to rear. Introduction of new species, intentionally or unintentionally, is one of the major threats to biodiversity today (Crémieux *et al.*, 2010). Responsible and sustainable introduction of a new livestock rearing system should therefore be based on species that pose no threat to the native ecosystem. The field cricket (*Teleogryllus testaceus*)

is a native species that inhabits paddy fields and fallow land in Cambodia. It is consumed by humans and a recent study show that it can be reared artificially (Megido *et al.*, 2016). Therefore *T. testaceus* is of high interest when examining the potential of cricket rearing systems in Cambodia.

It is known that the house cricket (*Acheta domesticus*) can survive and grow well on a variety of organic materials (Makkar *et al.*, 2014) including forage diets (Tyree *et al.*, 1974). The study by Megido *et al.* (2016) indicate that *T. testaceus* might have similar capacity. Accordingly, agricultural and food industry by-products and plants considered as weeds are potential cheap and sustainable feed sources. In Cambodia, agricultural by-products such as rice bran and cassava tops or leaves are potential feed resources. By-products from the food industry, such as spent grain and residues from mungbean sprout production, may also have potential. Some weeds are already harvested and used as feed for livestock. For example, *Commelina benghalensis* and *Boerhavia diffusa* can be used to feed pigs (Chikwanha *et al.*, 2007) and *Synedrella nodiflora* and *Amaranthus spinosus* have been used as feed and food (Adjibode *et al.*, 2015). Moreover, *Cleome viscosa* has been suggested to be useful as a feed for ruminants (Akinfemi and Mako, 2012). All the plant species discussed above are present in Cambodia and can be gathered and/or acquired free of charge.

The aim of this study was to evaluate survival and growth of Cambodian field cricket fed a set of local weed species and agricultural and food industry by-products. The hypothesis was that some of the materials tested have potential to be used as feed. However, since this cricket species is not commercially available, an initial challenge was to capture wild individuals and to rear and produce new generations from them, in order to provide the experiment with nymphs.

## 2. Materials and methods

### Capture and rearing

Males and females (ratio estimated to approximately 50:50) of *T. testaceus* were caught by hand in July 2014 from banks of water channels adjacent to rice fields at two locations in Cambodia, a site in the province of Kampong Thom (162 km Northeast of Phnom Penh, latitude 12.516782, longitude 105.111319) and a site in the province of Kampong Chhnang (100 km North of Phnom Penh, latitude 12.215312, longitude 104.673257). A 2 kg sample of crickets was caught at each location, corresponding to approximately 2,000 individuals/location. The crickets were transported by car (4 h at <30 °C) to the research farm of the Center for Livestock and Agricultural Development (CelAgrid) in Phnom Penh, Cambodia (latitude 11.442789, longitude 104.877337). During transport, the crickets were held in plastic cages (60×42×31 cm) with 0.5 kg crickets (~500

individuals) per cage. All cages contained conventional cardboard egg holders to provide hiding places and shadowy areas for the insects.

At the farm, the crickets were moved to concrete pens (width 2 m, length 3 m, height 0.7–1.0 m) covered by 1 mm mesh net and with a 10 cm water channel around to protect them from ants and predators. The density was 700–950 crickets per m<sup>3</sup>. The crickets had free access to chicken feed (Top Feed, Pathum Thani, Thailand) and water on plates (511 cm<sup>2</sup>, depth 1 cm, height 1.5 cm, 1 plate per m<sup>2</sup>). Stones were added to the plates to aid drinking and reduce the risk of drowning. Cardboard egg holders were placed in the pens as hiding and moulting places for the crickets. Plastic bowls (24 cm<sup>2</sup>, depth 6.5 cm; 4 bowls per m<sup>2</sup>) containing biochar were placed inside the pens as a substrate for egg laying. The crickets started to lay eggs within two weeks after capture. Hatching of the second generation was allowed from bowls presented to the crickets for 48 h (to minimise the variation in hatching time). The second generation crickets were kept in the same way as the first, except for a higher density (~1,400 crickets/m<sup>3</sup>), and reached maturity and laid eggs within three months after hatching. The pens were cleaned between each generation. The third generation of crickets comprised approximately 24,000 individuals. Individuals at the age of seven days from this generation were used in the feeding experiments. Prior to removal, the nymphs were fed chicken feed.

### Experimental design, preparation and management

The effect on survival and growth of field crickets fed agricultural and food by-products and weeds was studied in a 70-day feeding experiment. First larval stage nymphs (7 days after hatching) that looked healthy and had normal morphology (e.g. no legs missing) and phenotype (i.e. body size within normal range of the age group) were selected. Each treatment was represented by four replicates and 20 crickets were randomly allocated to each replicate. All individuals in a replicate were weighed together using an electronic scale (model TAJ202; Ohaus, New York, NY, USA; capacity: 200 g × 0.01 g). There was no difference in initial mean weight per individual between treatments (0.013±0.008 g, *P*>0.05 using ANOVA). The replicates were housed in plastic cages (60×42×31 cm<sup>3</sup>). A hole (36×9 cm<sup>2</sup>) was made in the lid of each cage and covered by a net (1 mm mesh) to allow air exchange. Water was offered on plates with stones (8 cm diameter, 2 cm depth) and plates were cleaned every two days. Apart from the water and the feed offered (see below), no other materials were present in the cages, to make sure that these were not eaten and hence used as a source of nutrients. Cages were cleaned every seventh day. The cages and the areas nearby were checked twice a day for spiders and other predators. The cages were set on wooden stands kept in water bowls to prevent ants from entering the cages.

The experiment was conducted between October 2014 and December 2014, which is early in the dry season. Temperature and humidity were recorded (model A103; Le Weeks, Nanjing, China) daily in three random cages. Mean temperature over the experimental period was  $29.0 \pm 1.7$  °C and humidity  $69 \pm 5\%$  and there were no differences between cages during the experimental period (ANOVA,  $P=0.71$  for temperature,  $P=0.69$  for humidity). The experiments were conducted under a 12-h natural daylight regime. At the end of the experiment, crickets were killed by soaking in fresh water.

### Feeds and feeding

In international commercial cricket rearing (Hanboonsong *et al.*, 2013; Lundy and Parrella, 2014) and in scientific studies (Megido *et al.*, 2016; Nakagaki and DeFoliart, 1991), crickets are commonly fed chicken feed. We therefore used a common chicken feed (Top Feed) as the control feed in this study. According to the manufacturer, the feed was composed of maize, broken rice, extracted oil, rice bran, fishmeal and soybean, and contained 21% crude protein (CP), 4% crude fat, 5% ash and 87% dry matter (DM). The experimental treatments comprised single species or product diets based on the following agricultural by-products: rice bran (RiceB), cassava plant tops (CassaT), water spinach (WateS) and spent grain (SpenG); waste from mungbean sprout production (Mung), and the plant weeds: *A. sessilis*, *A. spinosus*, *C. benghalensis*, *Cleome rutidosperma*, *C. viscosa*, *B. diffusa* and *S. nodiflora*.

The chicken feed, RiceB and SpenG were bought from local markets. Mung was received free from a local mungbean sprout producer. Chicken feed and RiceB were stored in a dry area during the experimental period. The fresh SpenG and Mung were collected from the producer every two weeks and stored in a refrigerator at 10 °C. WateS was harvested in the fields around the station and CassaT was harvested from a farm about 7 km from CelAgrid. The WateS and CassaT were collected once a week and cleaned, washed, trimmed and bunched before being stored at 10 °C. *S. nodiflora* (grows in upland fields and wastelands) was collected once every two weeks by a farmer at Kompong Sela, Koh Kong province, in the south of Cambodia, transported by car (about 4 h) to CelAgrid and prepared (cleaned, washed, trimmed and bunched) and stored at 10 °C. *A. sessilis*, *A. spinosus*, *C. benghalensis*, *C. rutidosperma*, *C. viscosa* and *B. diffusa* are common weeds found on the banks of streams and ponds, in paddies, shallow waterways, upland fields, roadsides and in wasteland throughout Cambodia. These weeds were collected mainly on roadsides and fallow land around CelAgrid once a week during the wet season (June–December) and prepared (cleaned, washed, trimmed and bunched) and stored at 10 °C.

The crickets were offered feed in the morning every second day. Chicken feed, RiceB, SpenG and Mung were offered *ad libitum* on small plastic plates. CassaT, WateS, *S. nodiflora*, *A. sessilis*, *A. spinosus*, *C. benghalensis*, *C. rutidosperma*, *C. viscosa* and *B. diffusa* were also offered *ad libitum*, but were laid in the bottom of the cages.

### Survival and weight data

Once a week, the number of live crickets was counted and the weight of the replicates was recorded. Survival rate in a replicate was calculated by dividing the number of live crickets at each time point by the initial number of crickets ( $n=20$ ). Individual cricket weight for each replicate was calculated by dividing replicate weight by the number of live crickets at the time of measurement. At day 42, crickets in last instar stage was observed and weight gain and feed conversion were therefore calculated at day 49 and at the end of the study (day 70). The size of *T. testaceus* is normally of commercial interest (i.e. individuals in last instar stage and females filled with eggs) around the age of 49 days. Mean individual weight gain was calculated by subtracting the mean individual weight at the start (day 0) from the mean individual weight at day 49 or 70. The amount of feed offered and rejected was recorded every two days. Mean feed consumption per week was calculated on DM basis (see below) by subtracting the weekly refusals from the feed offered and dividing this by the mean number of crickets alive at the time of measurement and the number of crickets in the previous week. Feed conversion ratio (FCR) was calculated by dividing the total mean individual feed consumption by the total mean individual weight gain. FCR was calculated only for feeds where survival and mean cricket weight were higher than 30% and 0.1 g, respectively, at day 42.

### Chemical analysis of feeds and refusals

Samples of feed and feed refusals were collected weekly. DM content of feed and refusals was measured by oven drying at 70 °C for 48 h until constant weight was achieved (Pen *et al.*, 2013). Analysis of nitrogen (N) was performed by the Kjeldahl method, and ash was determined by combustion in a muffle furnace at 550 °C for 4.5 h (AOAC, 1990). CP content was calculated as  $N \times 6.25$  and organic matter as DM less ash. Crude fat and crude fibre (CF) were analysed according to AOAC (1990) and gross energy (GE; MJ/kg DM) was calculated as  $17.6 + 0.0617 \text{ CP} + 0.2193 \text{ EE} + 0.0387 \text{ CF} - 0.1867 \text{ ash}$  (Sauvant *et al.*, 2004).

### Statistical analysis

Normal distribution of data was verified by residual plots and weight data had to be log-transformed. ANOVA was performed using the R software (version 3.2.2; <https://www.r-project.org/>). The model  $y_{ijk} = \mu + a_i + \beta_j + \tau_k + \beta\tau_{jk} + e_{ijk}$



was used for weight data, where  $y_{ijk}$  is the observation,  $\mu$  the general mean value,  $a_i$  the random effect of replicate  $i$ ,  $\beta_j$  the effect of treatment  $j$ ,  $\tau_k$  the effect of time point  $k$ ,  $\beta\tau_{jk}$  an interaction between treatments (feed types) and time (week), allowing for different time developments for the different treatments and  $e_{ijk}$  the residual. A similar model: ( $y = XB + Zu + e$ , a logistic mixed model) was used for the survival data, where  $y$  is a vector that contains the responses for all individuals stacked on top of each other,  $X$  is a design matrix,  $B$  is a parameter vector (as in general linear models, a random effect),  $u$  is a vector of random effects,  $Z$  is a design matrix for the random effects and  $e$  is a vector of residuals. The residuals were assumed to be correlated within replicates according to an autoregressive AR(1) model. The generalised linear mixed model was used for the proportions  $x/n$ , where  $x$  is the number of survivors at different time points of weeks and  $n$  is the number of individuals at the start. Both models were analysed using the GLS (generalised least squares) function from package NLME (includes the repeated structure, i.e. observations repeated within replicate).

Weight gain and feed consumption were analysed using one-way ANOVA (aov command for comparisons between feeds). Comparison with the control (chicken feed) was made by using the glht() command and a Tukey contrast by package multcomp. Data on starting weights and cage temperature and humidity during the experiment were analysed using ANOVA and a Tukey contrast (models including for weights: treatment and replicate and for temperature and humidity: day and cage (commands lm() and lsmeans()). Feed conversion rate was analysed using

one-way ANOVA and Dunnett multiple comparisons with a control in Minitab (Minitab Inc., State College, PA, USA). Proc corr in SAS (package 9.4; SAS Institute Inc., Cary, NC, USA) was used to calculate correlations between weight gain and energy and nutrient contents of the feeds. The values presented are least square means  $\pm$  standard error. The level of statistical significance was set to  $P < 0.05$ .

### 3. Results

#### Chemical composition of feeds

Energy content in the feeds ranged from 16.5 to 20.2 MJ GE/kg DM and the CP content ranged from 10 to 29% of DM. The lowest and highest energy content were observed in *C. benghalensis* and spent grain, respectively, and the lowest and highest CP content in RiceB and CassaT, respectively (Table 1).

#### Survival rate

Overall, survival was not different from chicken feed with the exception of crickets fed *B. diffusa*, which had lower survival ( $P < 0.05$ ) from day 14 to day 49 (Figure 1). From day 35 to day 49, survival on *A. sessilis* was also lower ( $P < 0.05$ ) than on chicken feed.

#### Weight

The change in body weight throughout the study is shown in Figure 2. There was no difference ( $P > 0.05$ ) in weight between crickets fed CassaT, *C. rutidosperma* and chicken feed throughout the study. From day 35 and throughout the study,

**Table 1. Dry matter content, chemical composition and estimated gross energy content of chicken feed, weeds, agricultural and food industry by-products used in a 70-day feeding study on local Cambodian field crickets (*Teleogryllus testaceus*).**

	Dry matter	Crude protein	Ash	Crude fibre	Crude fat	Gross energy
Feeds	%	% of dry matter				MJ/kg DM
Chicken feed <sup>1</sup>	91.1	23.4	6.5	5.7	9.0	20.0
<i>Alternanthera sessilis</i>	19.5	15.0	13.5	25.4	5.6	18.2
<i>Amaranthus spinosus</i>	14.4	24.4	21.5	16.9	6.0	17.1
<i>Boerhavia diffusa</i>	14.1	23.8	20.4	14.0	4.7	16.8
<i>Cleome rutidosperma</i>	14.3	22.2	17.3	28.3	3.2	17.5
<i>Commelina benghalensis</i>	9.1	16.7	19.6	19.6	3.7	16.5
<i>Synedrella nodiflora</i>	11.3	19.3	16.5	21.0	4.8	17.6
<i>Cleome viscosa</i>	13.9	22.9	17.5	22.0	2.5	17.1
Cassava tops	19.7	28.6	5.2	14.2	4.6	20.0
Rice bran	92.0	10.2	11.9	23.0	12.0	19.5
Water spinach	10.0	23.5	11.9	13.2	3.4	18.1
Mungbean sprout residue	20.4	19.4	3.9	28.9	2.2	19.7
Spent grain	24.9	27.1	3.0	16.0	4.1	20.2

<sup>1</sup> Top Feed, Pathum Thani, Thailand.

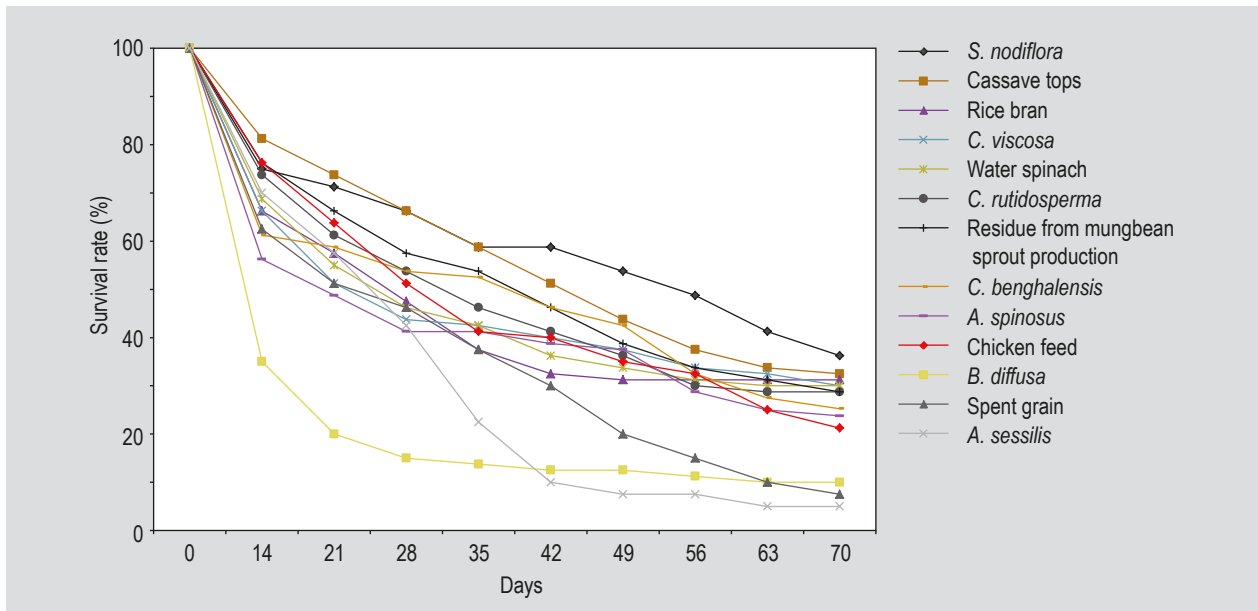


Figure 1. Survival in crickets fed chicken feed or one of twelve weeds, agricultural and food industry by-products for 70 days. The twelve treatments were single product diets. Standard error was for cassava tops 5.6, chicken feed 5.7, *Cleome rutidosperma* 5.0, *Cleome viscosa* 3.5, *Synedrella nodiflora* 4.1, residue from mungbean sprout production 5.2, *Commelina benghalensis* 4.2, spent grain 6.1, water spinach 4.2, rice bran 4.2, *Boerhavia diffusa* 2.5, *Alternanthera sessilis* 8.1 and *Amaranthus spinosus* 3.4.

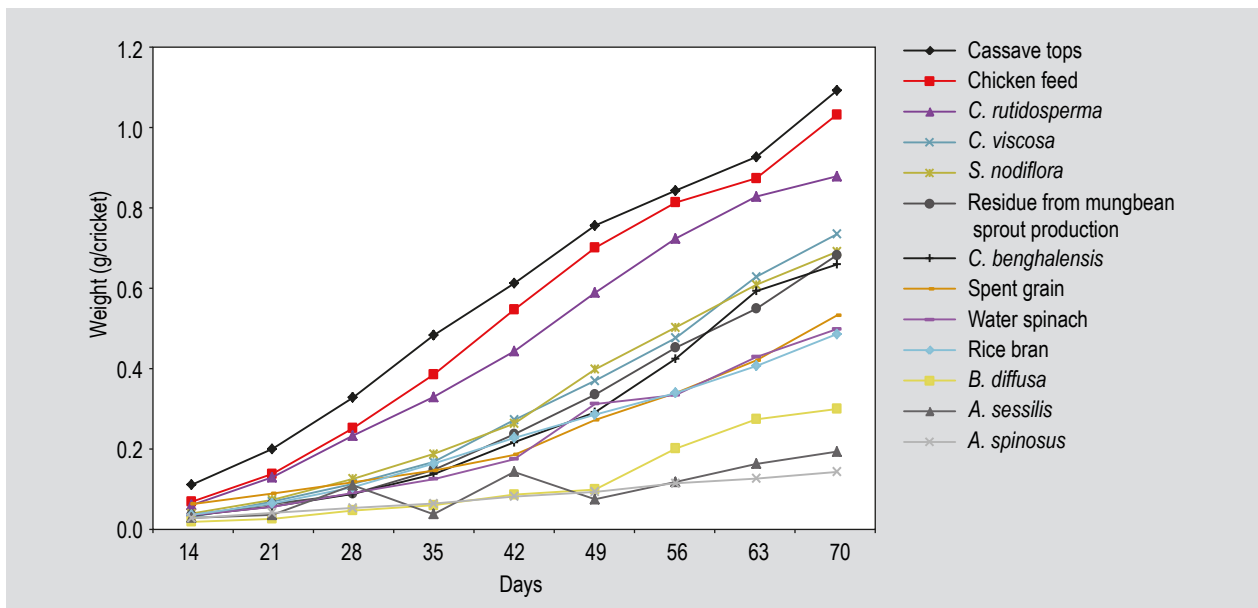


Figure 2. Weight of crickets fed chicken feed or one of twelve weeds, agricultural and food industry by-products for 70 days. The twelve treatments were single product diets. Standard error was for cassava tops 0.11, chicken feed 0.11, *Cleome rutidosperma* 0.09, *Cleome viscosa* 0.08, *Synedrella nodiflora* 0.07, residue from mungbean sprout production 0.07, *Commelina benghalensis* 0.07, spent grain 0.05, water spinach 0.05, rice bran 0.05, *Boerhavia diffusa* 0.03, *Alternanthera sessilis* 0.02 and *Amaranthus spinosus* 0.01.

the weight was lower in crickets fed *C. rutidosperma*, Mung, SpenG and *S. nodiflora* compared with crickets fed chicken feed, with the exception of day 63 when the weight of crickets fed *C. rutidosperma* and *S. nodiflora* was not different from that of crickets fed chicken feed (Figure 2). From day 28 and until 70, the weight of crickets fed *C. benghalensis*, RiceB and

WateS was lower ( $P < 0.05$ ) than that of crickets fed chicken feed with the exception of day 63, when *C. benghalensis* was not different from chicken feed (Figure 2). For crickets fed *A. sessilis*, *A. spinosus* and *B. diffusa* the weight was lower ( $P < 0.05$ ) than in crickets fed chicken feed already at day 21 and remained so until day 70 (Figure 2).

Feed consumption, weight gain and feed conversion

Compared with chicken feed, feed consumption at day 49 was lower for WateS and all weeds except *C. rutidosperma* (Table 2). Weight gain at day 49 was lower in crickets fed WateS, *C. benghalensis*, Mung and RiceB compared with those fed chicken feed (Table 2). There were no differences in FCR at day 42 except for RiceB, where FCR was higher than for chicken feed (Table 2).

There were no differences in feed consumption and FCR after 70 days, but weight gain was lower than with chicken feed for all test feeds except *C. rutidosperma* and CassaT (Table 2). In SpenG, *A. sessilis*, *A. spinosus* and *B. diffusa*, survival or mean weight at day 42 was less than 30% and 0.1 g, respectively, and FCR was therefore not calculated. There were no correlations between weight gain and feed CP, ash, crude fibre, crude fat and gross energy content.

4. Discussion

To our knowledge, this is the first study showing that it is possible to rear Cambodian field crickets (*T. testaceus*), using simple means, on local weeds, agricultural and food industry by-products. Interestingly, crickets fed cassava plant tops and *C. rutidosperma* had the same growth performance as crickets fed chicken feed and the use of these feeds in a local rearing system could therefore be an option for smallholder farmers and low income families. Cassava tops may be superior to cassava leaves as feed for *T. testaceus*, since cassava leaves have been shown to result in lower body weight than chicken feed (Megido *et al.*, 2016). Despite the lower growth performance observed for some of the other materials evaluated in the present study, most can also be used as cricket feed if resources are scarce. A combination

of different weeds and by-products might also increase the performance compared with the outcome in the present study, since potential deficiencies and imbalances might be rectified. This is supported by observations by Megido *et al.* (2016), who studied mixed diets. A successful combination already identified is cassava leaves and chicken feed, which have been shown to increase total and daily egg production and extend the laying period in three cricket species compared with feeding chicken feed and papaya leaves (Fuah *et al.*, 2015). However, the relevance of that diet for growth and survival of cricket nymphs remains to be investigated.

The present study also indicated that *A. sessilis*, *A. spinosus* and *B. diffusa* should not be used as feed for *T. testaceus*, since survival and growth were extremely low. All weeds had comparatively high ash content, and thus mineral imbalances might have been present and affected survival and growth. Further studies are needed to confirm this. Another issue with the use of weeds collected from roadsides and fallow lands might be contamination with various substances like heavy metals or insect eggs (that could not be observed by the eye). In this study, weeds were washed 2-3 times with fresh water before feeding to limit intake of unwanted substances attached to the surface. However, if substances are accumulated within the plants this treatment will not affect this, and food safety could possibly be threatened. Further studies are therefore needed on food safety aspects of collecting plants at places where contamination could be a risk.

However for all feeds, including chicken feed, the survival rate was lower than 70% already after 28 days and continued to decrease throughout the study. This survival rate is lower than values reported elsewhere for other reared cricket species, which range from 76 to 100% at the age of 3-4 weeks

Table 2. Total mean feed consumption (FC, g/cricket), weight gain (WG, g/cricket) and feed conversion ratio (FCR, g/g) in crickets (*Teleogryllus testaceus*) fed chicken feed, weeds, agricultural and food industry by-products for 49 and 70 days.<sup>1</sup>

Feed	FC 49 d	WG 49 d	FCR 49 d	FC 70 d	WG 70 d	FCR 70 d
Chicken feed <sup>2</sup>	1.21	0.59	1.85	2.05	1.01	2.25
<i>Cleome rutidosperma</i>	1.02	0.53	1.90	2.10	0.86	2.35
<i>Cleome viscosa</i>	0.69*	0.36	2.00	1.99	0.72*	2.98
<i>Commelina benghalensis</i>	0.53**	0.28*	2.03	1.83	0.65**	3.40
<i>Synedrella nodiflora</i>	0.61**	0.35	1.68	1.07	0.68**	1.70
Mungbean sprout waste	1.12	0.31*	3.68	1.97	0.67**	3.23
Cassava tops	1.09	0.75	1.60	2.14	1.06	2.05
Rice bran	0.75	0.26**	3.88*	1.44	0.47***	3.90
Water spinach	0.65**	0.30*	2.33	1.32	0.49***	2.80
Standard error	0.18	0.09	0.66	0.55	0.10	1.22

<sup>1</sup> \* *P*<0.05; \*\* *P*<0.01; \*\*\* *P*<0.001 in comparison with chicken feed.

<sup>2</sup> Top Feed, Pathum Thani, Thailand.

(Clifford *et al.*, 1977; Neville and Luckey, 1962; Tyree *et al.*, 1974). It is also lower than values recently reported for *T. testaceus* (Megido *et al.*, 2016). However, in that study the individuals were heavier than in ours, indicating that they might have been older at the start of the experiments. The reason for the low survival in our study is unclear. Since this is the first time this species has been studied in captivity, knowledge on their specific requirements is limited. A comparatively high mortality could be expected in the first generations when wild species adapt to rearing circumstances. However, ambient temperature is one critical factor for survival. In another cricket species, *A. domesticus*, the ideal temperature appears to be around 30 °C (Clifford and Woodring, 1990; Makkar *et al.* 2014; Roe *et al.*, 1980). If that is relevant also for *T. testaceus*, the rearing conditions we provided were optimal with respect to mean temperature and could not have caused the high mortality. However, according to Clifford *et al.* (1977), small temperature changes can have drastic effects on the life cycle duration of crickets. This is also in accordance with observations by Lachenicht *et al.* (2010). They examined the effects of 7-9 days acclimatisation to temperatures 4 °C above and below a target temperature of 29 °C and found that temperature variations increased mortality, and especially high temperature acclimation resulted in a dramatic increase in mortality. In that study, mortality was 60-70% in crickets kept at 33 °C. In the present study, crickets were not reared in a room with strict climate control and it is therefore possible that variations in ambient temperature could have increased mortality.

Studies on the effect of ambient humidity on survival of crickets in captivity are scarce. According to Ghouri and McFarlane (1958) and Clifford and Woodring (1990), humidity should be below 50% for *A. domesticus*. A study of wild populations of field crickets in mung bean crops in Pakistan (Khan *et al.*, 2011) also indicated that humidity is of significant importance. That study showed that population size reached its maximum when the crop was in its initial stages, temperature was 33 °C and humidity was 50%, but also that there was a significant negative correlation between the amount of crickets and relative humidity. Therefore, it is possible that the high humidity (around 70%) in our study could have increased mortality. Factors such as water availability, density of crickets (Clifford and Woodring, 1990; Clifford *et al.*, 1977) and species strain (Ghouri and McFarlane, 1958) may also affect life cycle duration, but further studies are needed to evaluate how this affects *T. testaceus*. Interestingly, the hydrogen cyanide (poisonous) and tannins (anti-nutritional) most likely present in the fresh cassava tops (Awoyinka *et al.*, 1995) had no apparent negative effect on the crickets in the present study. There should be no potential hazards for people consuming crickets reared on cassava tops since hydrogen cyanide seems not to be accumulated or stored in blood and tissues (Simeonova and Fishbein, 2004).

About 30% of the crickets fed cassava tops, *C. rutidosperma* and chicken feed reached last instar stage at 42 days of age and at an average body weight in the range 0.33-0.48 g. Similar weights have been reported for *A. domesticus* (0.37-0.48 g) at the same development stage (Clifford and Woodring, 1990; Nakagaki and DeFoliart, 1991; Patton, 1963). Although there were considerable variations in nutrient content between the feeds tested in this study, there was no simple relationship between nutrient content and weight gain. For example, CP content of spent grain was similar to that of cassava tops, but growth performance was still low for spent grain. If there are no major differences in digestibility, mineral content or content of anti-nutritional substances between these feeds, the difference is most likely explained by the amino acid profile. Nakagaki and DeFoliart (1991) also found that there were no differences in weight of crickets fed a 14% CP diet (commercial non-breeding rabbit feed, 20% crude fibre) for 21 days compared with crickets fed a 22% CP diet (corn and soybean-meal based chicken diet, <5% crude fibre). Moreover, Megido *et al.* (2016) found no correlation between dietary CP content and CP content of *T. testaceus*. However, further studies are needed to better understand the protein and amino acid requirements of crickets and the impact of dietary chemical composition on cricket composition.

Crickets in the present study did not seem to respond negatively to high crude fibre content in the feed, since the content was high in *C. rutidosperma* and, despite this, cricket weight still remained among the highest and feed conversion rates among the lowest. In *A. domesticus*, it has been shown that growth is promoted by inclusion of roughage carbohydrates in the diet, but also that carbohydrates such as D-ribose, D-xylose, D- and L-arabinose and L-sorbose are inhibitory to growth (Neville and Luckey, 1962).

The FCR ranged from 1.6 to 3.9 at day 49. In chicken feed, cassava tops and *C. rutidosperma*, it was  $\leq 1.9$ , which is similar to the FCR reported for *A. domesticus* fed chicken feed and food waste (1.3-1.8) (Collavo *et al.*, 2005; Lundy and Parrella, 2014). Since the FCR at day 49 (when the size of crickets is of commercial interest) was the same for chicken feed, cassava tops and *C. rutidosperma*, use of the latter two as feed for crickets is an interesting option from an economic perspective, since they are free of charge. However, if paid labour is used to collect the feeds or if the demand of the plant material increases the situation will be different.

In conclusion, this study shows that it is possible, using very simple means, to rear local field crickets at ambient temperature in Cambodia. Cassava tops and *C. rutidosperma* both have great potential to be used as feed for field crickets. Most of the other weeds, agricultural and food industry by-products tested here also have potential for use as cricket feed, alone or in combination. Exceptions were the weeds *A. sessilis*, *A. spinosus* and *B. diffusa*, which cannot be recommended for feed use.



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