Chapter

Mixed Diets Enhance Edible Grasshopper, *Ruspolia differens* (Orthoptera: Tettigoniidae) Performance during Mass Rearing

Geoffrey Maxwell Malinga, Robert Opoke and Karlmax Rutaro

Abstract

Mixing of diets is a notable dietary practice that is believed to improve performance-related characteristics such as growth, survival rate and egg-laying potential among insect herbivores. However, currently there is limited information regarding the performance of edible insects either on artificial and natural diets or their mixtures. This chapter reviewed recent literature on performance of a seasonally harvested and a widely consumed edible grasshopper, *Ruspolia differens* (Orthoptera: Tettigoniidae) reared on various artificial and natural diets. Our aim is to highlight diets and diet mixtures that results in the highest *R. differens* production. The results of the review show that *R. differens* performs better on mixed diets than on single or less diversified diets. In all reviewed studies, edible grasshoppers fed mixed diets either of natural plants or artificial diets achieved highest final weights, highest survival, highest fecundity and fastest development times than less diversified diets. The information is useful in designing technologies for large-scale rearing program for this species.

Keywords: insect farming, developmental time, captive rearing, African edible bush cricket

1. Introduction

Diet mixing, i.e., feeding on more than one plant species or food resource is a well-known feeding habit among polyphagous insect herbivores and is believed to be associated with increased growth, shorter development time, survival rate and fecundity [1, 2]. This is because mixed diets could allow nutrient complementation and reduce the amounts of toxins in solitary foods (reviewed in [3, 4]). Several polyphagous orthopteran species have been shown to enhance their performance by diet mixing and exhibit higher fitness and survival rates, and faster growth on mixed diets than on single resource diets [1, 2, 5–7]. However, despite this widespread evidence of diet mixing in insect herbivores, in contrast, very little has been done to review the effect of diet mixtures on the performance of edible insects, which hamper the development of a suitable diet or diet mixtures for sustaining mass-rearing.

The edible grasshopper, *Ruspolia differens* (Orthoptera: Tettigoniidae) is a seasonally harvested edible insect species. This edible grasshopper species is largely found in tropical Africa particularly in Kenya, Angola, Uganda, South Africa, Ghana, Central African Republic, Democratic Republic of Congo, Zambia, Rwanda, Burundi, Mauritius, Madagascar, Ivory Coast, Tanzania, where it is eaten as a delicious food [8–15]. It is nutritionally rich and healthy. *Ruspolia differens* have been found to have high levels of protein (34-73%), fat content (33-48%), carbohydrates (3-6%), polyunsaturated fatty acids (89% of lipids), chitin (10-15%) and several minerals including phosphorous (141-673 mg/100 g), potassium (446-673 mg/100 g), calcium (34.9-128 mg/100 g), Fe (17 mg/100 g) and zinc (12-17 mg/100 g). This katydid grasshopper is predominantly nocturnal, showing distinctive colour polymorphism and sex dimorphism [8]. There are six colourmorphs comprising of green, purple-stripped green, purple-suffused green, brown, purple-stripped brown and purple-suffused-brown [16] although the green and brown colourmorphs tend to dominate. *Ruspolia differens* is characterised by a unique swarming phase that exist during rainy seasons and a non-swarming phase (also called local population) whose developmental stages are found in the field all over the year [15, 17]. In East Africa, swarming occurs twice annually in April–May and November–December [8], and the source populations of the swarming individuals are believed to originate locally [17]. Male adults are smaller than females, and are characterised by longer antenna and possess a pair of tongue-like metathoracic flaps and dorsoventrally bi-lobed cerci, while, females are identified by the presence of a long slender ovipositor [10, 15, 16, 18–21]. The life cycle is variable but roughly 147 days and there are six nymphal stages for male and seven for females [12]. Eggs are oviposited in the leaf sheaths of grasses, shoots of Panicum sp., maize seedlings and *Pennisetum sp*, and cotton wool and folded plastic cloth [22–24]. Hatching happens after 11-25 days and is determined by the moisture level [22, 24, 25]. *Ruspolia differens* is a facultatively oligophagous grass-feeder and can feed on a range of grass and sedge species [17, 26, 27] and various artificial diets [12, 25, 28]. However, the species is highly selective preferring inflorescences over leaves [27] but they are flexible if a limited range of diets are available [27, 28]. Despite the large number of recent research works on *R. differens*, however, there is dearth of information regarding the performance of edible insects either on artificial and natural diets or their mixtures. This chapter systematically reviewed all recent peer-reviewed literature on the development and reproductive performance of edible grasshopper, *Ruspolia differens* (Orthoptera: Tettigoniidae) reared on various artificial and natural diets over the last 20 years. Our aim is to highlight diets and diet mixtures that results in the highest *R. differens* production. The information is useful in designing large-scale rearing program for this species.

2. Materials and methods

A comprehensive literature search was conducted within the electronic databases such as Scopus, Web of Science and Google Scholar to identify peer-reviewed journal articles focussing on performance of *R. differens* on mixed or diversifying diets. In the search, following key words were used to demarcate the species in the search: *Ruspolia differens*, Long-horned grasshopper, Edible bush cricket, Swarming conehead, Mixed diets and Performance. The inclusion criteria included only peer-reviewed papers published within the last 20 years (from 2000 to 2020, the period of active research on *R. differens*). Because of the aggravated risk of bias as a result of translation of information from other languages to English, our search was confined to English language only. Dissertation, conference proceedings and papers published in other languages were excluded from the review. The title, abstracts and full texts of the articles were screened against the selection criteria. The search yielded six articles out of which three were relevant and were included in the systematic review. The statistical assessments of effects of dietary treatment on development time (days) from the first instar to adult and fresh weight at adult stage and female fecundity were done using Linear Mixed model with type III sum of squares (sex and diet-sex interaction included as fixed factors and block as a random variable) followed by Bonferroni post hoc pairwise comparisons. A logistic regression model was used to determine whether diet treatment (predictor variable) predicts the nymphal survival to adulthood (outcome variable) with block included as a random variable. All statistical analyses were conducted using IBM SPSS Statistics software version 26. For artificial diets study, Linear contrasts within the analysis of variance (ANOVA) were employed to examine the existence of a linear trend in the insect performance variables along the food resource diversity gradient.

3. Results

The reviewed articles were based on recent studies conducted over the last 20 years (2000 to 2020) in Uganda and Kenya. Most of these articles were published in 2018. The studies focused on identification of articles that focused on performance of edible grasshopper, *R. differens* on either natural or artificial diets.

3.1 Performance of *R. differens* on mixed artificial diets

The development of a successful and sustainable mass rearing strategy requires knowledge of potentially suitable diet mixtures which can best support the insect's growth, development and survival. A study by Malinga et al. [25] in Uganda, examined how diet treatments representing a gradient of increasing food resource diversity, ranging from one food (single-food treatment) to mixtures of two, three, five, six and eight foods (**Table 1**), affect the developmental and reproductive performance of the edible grasshopper, *R. differens*. In all reviewed studies, approximately equal amounts of foods were administered ad-libitum to each individual in the diet mixture till moulting to adult, and water was provided either by introduction of moistened tissue paper or by spraying droplets of water with a portable hand sprayer.

More diversified diets resulted in shorter development time rather than in the single diet or less diversified diets and there was a significant decreasing linear trend in the development time along the food resource diversity gradient, indicating that as food resource diversity increases from single to more diversified diet mixtures, development time decreases proportionally. The improved performance are likely due to the utilisation of a balance of high-quality nutrients in mixed diets [2]. Benefits of diet mixing on insect performance (e.g., developmental time) has previously been documented also on several generalist Acrididae grasshoppers [1–3, 5, 29]. These studies have found generalist grasshopper to perform better on high quality foods which in turn determines their growth and developmental rate [30]. Increasing dietary diversification resulted in greater adult weight and there was a significant increasing linear trend in the adult fresh weight along the food resource diversity gradient. Furthermore, the experimental results of this work indicated that dietary mixing greatly improved female fecundity (total number of eggs laid per female, on average roughly 90-140 eggs). There was also a significant increasing linear trend in the female fecundity along the food resource gradient. Improved

Treatment code	Treatment name	Composition
One	Single food treatment	Rice seed head
Two	Two food mixtures	Rice seed head
		Finger millet seed head
Three	Three food mixtures	Rice seed head
		Finger millet seed head
		Wheat bran
Five	Five food mixtures	Rice seed head
		Finger millet seed head Wheat bran
		Chicken superfeed egg booster (from super Feeds Farm Products Ltd., Kampala, Uganda)
		Sorghum seed head
Six	Six food mixtures	Rice seed head
		Finger millet seed head Wheat bran Chicken superfeed egg booster Sorghum seed head Germinated finger millet
Eight	Eight food mixtures	Rice seed head
		Finger millet seed head
		Wheat bran
		Chicken superfeed egg booster Sorghum seed head Germinated finger millet
		Simsim cake
		Dog biscuit pellet

Table 1.
Compositions of diets used in the performance experiments.

fecundity in mixed diets has also been reported for the generalist grasshoppers *Parapodisma subastris* (Huang) [1] and *Chorthippus parallelus* (Zetterstedt) [2] (Orthoptera: Acrididae).

Finally, more diversified diets resulted in greatly improved survival rates (approximately 31-51%) of *R. differens* to adulthood than when reared on single resource diets (11%). Even with a minimal diet diversification, nymphal survival to adult was greatly improved. The odds of survival (the probability of surviving relative to dying) was four to eight times higher for individuals fed mixtures of artificial foods than in those fed one food only. Numerous studies have shown that enhanced survival and development in the majority of herbivorous insects correspond to diet quality and quantity on which the insects are fed [30–32]. For instance, Oonincx et al. [32] established that larvae of the black soldier fly, *Hermetia illucens* (L.) (Diptera: Stratiomyidae) reared on vegetable by-products diets high in protein achieved a relatively shorter development time (21 days) that their counterparts reared on low-protein diets (37 days). Thus, the remarkably lower survival observed on single than in mixed diets could reflect the low protein and high starch content in solitary foods, as also found previously with mealworms [29]. The results of our review demonstrate how a cost-effective optimal mass production of *R. differens* could be achieved even with minimal dietary mixtures.

3.2 *Ruspolia differens* performance on diet mixtures of natural food plants

In a study evaluating the suitability of inflorescences for rearing *R. differens* [26], individual *R. differens* were reared from first instar to adults on diet treatments representing a gradient of five diversifying dietary mixtures of host plant species ranging from one to mixtures of two, three, five and seven species (**Table 2**). The gradient represented the hierarchy of use, based on a survey of potential food plants of *R. differens* used in the field in Uganda [17], whereby *Brachiaria ruziziensis* was the most used and *B. ruziziensis* and *P. maximum* were the two most used host plants in the field, respectively.

Treatment code	Treatment name	Composition
One	One host plant	*Brachiaria ruziziensis*
Two	Two host plants mixture	*B. ruziziensis*, *Panicum maximum*
Three	Three host plants mixture	*B. ruziziensis*, *P. maximum*, *Hyparrhenia rufa*
Five	Five host plants mixture	*B. ruziziensis*, *P. maximum*, *H. rufa*, *Chloris gayana*, *Cynodon dactylon*
Seven	Seven host plants mixture	*B. ruziziensis*, *P. maximum*, *H. rufa*, *Ch. gayana*, *Cy. dactylon*, *Sporobolus pyramidalis*, *Pennisetum purpureum*

Table 2.
Host plant combinations used in mixtures.

The rearing containers were placed in blocks, each containing one replicate of each host plant with one *R. differens* individual per container. In each container, approximately equal quantities of foods were randomly placed. The results showed that dietary mixture of grass species inflorescences is beneficial for the survival and for shortening the development time of the edible *R. differens* but not for achieving a higher adult emergence weight. For example, the nymphal developmental time was significantly shorter (on average 16 days shorter) when *R. differens* individuals were reared on more diverse diets than when reared on single diets (>90 days for diets with 1-2 plant species; <80 days for diets with 5-7 plants). Similarly, the survival to adult stage was higher in the most diversified diet mixture than in the least diversified diets, ranging from 12.5-15% in the three least diversified diet treatments to 40-65% in the two most diversified diets with an overall average nymphal survival of 30%. In another study conducted by Ssepuuya et al. [33], highest survival of *R. differens* was found after eight weeks on mixed diets comprising of green stems and leaves of three plants *C. dactylon*, *P. maximum* and *E. africana*) than in single plant diets. Consistent with our reviewed findings, previous works on generalist grasshoppers, *P. subastris* (Huang) [1] and *C. parallelus* (Zetterstedt) [2] also recorded highest survival in the 6-and 8-food plant (grass species) mixtures, respectively, than in food treatments with only a single plant species. The better performance on diet mixtures may be driven by a better gain of nutrients amounts in mixed than in single host plants [2].

4. Conclusion and future prospects

To conclude, the results of this review collectively provide evidence that *R. differens* performs better on mixed diets than on single or less diversified diets.

These results emphasise the importance of diet mixing in the optimisation of edible grasshopper, *R. differens*, production during mass-rearing programs. However, to upscale from current laboratory rearing to full-scale mass-rearing programmes suitable for local farmers or entrepreneurs, there are still several knowledge-gaps and steps remaining, e.g., what are the most cost-effective feeds in mass-rearing, and could side-streams from East-African food-industry or agricultural farms be included in feeds (reducing the costs and enhancing environmental sustainability of the rearing)? Do the used feeds in rearing modify the taste of *R. differens*? Economics of rearing: what feed and feeding regimes are economically viable?. Furthermore, research is needed to determine the combined effects of diet mixtures of natural and artificial foods on the performance of *R. differens*.

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