



# Assessment of the efficiency of local protein-rich products for mass rearing of the tephritid fruit fly *Bactrocera dorsalis* (Hendel) and its biocontrol agent *Fopius arisanus* (Sonan) in laboratory conditions

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## Abstract

*Fopius arisanus* (Sonan) is an ovo-pupal parasitoid used as biocontrol agent against the oriental fruit fly *Bactrocera dorsalis* (Hendel). The effectiveness of this parasitoid in the natural agro-ecosystems depends partly on the quality of its mass rearing system. Preparation of the imported hydrolysate yeast-based diet usually results in high costs as the hydrolysate yeast is very expensive. We therefore assessed some local protein-rich products including powders of moringa, cassava leaves, and soybean flour as substitutes to the expensive imported hydrolysate yeast. The results revealed that eggs hatchability of *B. dorsalis* on moringa diet was higher while the adult emergence rate was lower. Cassava leaves-based diet did not differ from the deactivated hydrolysate yeast diet in eggs hatchability, pupal recovery, and emergence rate of adult *B. dorsalis*. However, *F. arisanus*-parasitized *B. dorsalis* egg hatchability on moringa leaves- and cassava leaves-based diets were higher than on the other diets. Pupal recovery from cassava leaves-, papaya- and soybean flour-based diets were lower than from the control. Parameters such as larval duration, larval survival rate and sex ratio did not differ significantly between treatments for any of the two insect species. The emergence rate of *F. arisanus* did not differ from one treatment to another. The best cost-effective local protein-based diet, available for local use and which offered performances close to those of the control diet, was the cassava leaves-based diet.

**Keywords** Larval artificial diet · Cassava leaves-based diet · Mass rearing · Protein

## Introduction

Fruit flies of the Tephritidae family belong to the most damaging insect pests of fruits with over 4,000 species distributed throughout the world (Hardy 1997). Many of these fruit fly species are capable of infesting a wide range of commercial and native fruits and the extent of infestation and damage varies among host fruit species (Shinwari et al. 2015). Of the fruit fly species, *Bactrocera dorsalis* (Hendel) (Diptera: Tephritidae), is one of the most devastating

species (Lux et al. 2003a; Goergen et al. 2011). In Africa, the extent of damage along with the cost associated with the management of this notorious insect pest may induce over US\$ 2 billion of financial losses per year (Mutamiswa et al. 2021). Under such circumstances, the development of control methods that can sustainably manage the pest is needed. In fact, most of the promising management strategies initiated against fruit fly species in Africa include the use of attractants and population monitoring, male annihilation technique, sterile insect technique, biopesticides, biological control, field sanitation, and the integrated pest management (Ekesi et al. 2016). Of these management strategies, classical biological control has received much attention considering the exotic origin of this pest (Ekesi et al. 2006).

The complex of natural enemies of fruit flies gathers parasitoids belonging to the Braconidae family have frequently been used in biological control with successful results (Lux et al. 2003b; Vargas et al. 2007; Gnanvossou et al. 2016).

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*Fopius arisanus* (Sonan) (Hymenoptera: Braconidae), an egg/larval/pupal endoparasitoid is originated from South-east Asia and has been widely used in the management of invasive and locally endemic fruit fly species around the world (Rousse et al. 2005; Goncalves et al. 2017; Groth et al. 2017; Lane et al. 2018). *Fopius arisanus* is reared in egg-larva of *B. dorsalis* under laboratory conditions. Several studies conducted under laboratory conditions showed the preference of *F. arisanus* for the Oriental fruit fly *B. dorsalis*, the Mediterranean fruit fly *Ceratitis capitata* (Wiedmann) (Diptera: Tephritidae) and the mango fruit fly *Ceratitis cosyra* (Walker) (Diptera: Tephritidae) (Bautista and Harris 1996; Mohamed et al. 2010). The parasitism rates of *F. arisanus* ranged from 46 to 70% on *B. dorsalis* when reared on an artificial diet and on a wild mango *Irvingia gabonensis* (Aubry-Lecomte) Baill, respectively (Mohamed et al. 2010, 2017; Gnanvossou et al. 2016). However, one of the major constraints in the wide utilization of this parasitoid species is the use of adequate mass rearing system for the establishment of stable colony in laboratory. In fact, scaling up of laboratory rearing to mass-production of insects remains one of the most important challenges of fruit fly management programs since mass rearing is required to facilitate research and implementation of control tactics such as classical biological control. Ekesi and Mohamed (2011a) reported that factors such as artificial larval and adult diets as well as rearing conditions affect the quality and performance of the insects produced from mass-rearing, and therefore warrant further investigations. Several studies have assessed the impact of larval food quality on the performance of *B. dorsalis* larvae and adults (Chang 2009; Anato et al. 2017).

Among the constituents of larval diets, proteins are important nutrients for cellular growth and are considered as a vital component for fruit flies' egg development which occur in oocyte maturation to the vitellogenic stage (Shinwari et al. 2015). The amount of protein intake plays key role in the total egg laying capacity of females (Alamzeb et al. 2006). As sources of protein, the hydrolysate yeast (*Saccharomyces cerevisiae* Meyen) imported from the United States of America results in relatively high insect production costs depending on its availability and formulations (Morelli et al. 2012). It is used in most of the bio factories as protein source in adult diets (Morelli et al. 2012). The high costs associated with insect production correspond to larva production, due to the investments in ingredients for the larval diet. Dominguez Gordillo (1996) reported that larval diet accounts for 40–50% of mass-rearing plan investments. The findings by Silva Neto et al. (2011) reported up to US\$ 14.00 cost required to obtain 88.86 g of hydrolyzed protein needed for the production of one million *C. capitata* pupae in the bio-production facility suggesting the development

of other alternative source of protein. It is therefore important to find a cost-effective source of protein which not only makes a balance between costs and insect quality, but also is available for local use. The present study attempted to assess locally available protein-based products as protein sources in the larval diet of *B. dorsalis* used as host of the egg-pupa parasitoid *F. arisanus*. Specifically, it assesses the effect of Moringa leaves powder (*Moringa oleifera* Lam); cassava leaves powder (*Manihot esculenta* Crantz), soybean flour (*Glycine max* (L.) Merrill), mixed with papaya (*Carica papaya* L.) pulp as local protein-based products, to optimize mass rearing technique of *F. arisanus* under laboratory conditions.

## Materials and methods

### Insects rearing

The tephritid fruit fly *B. dorsalis* and the parasitoid *F. arisanus* used in this study were obtained from the Laboratory of Agricultural Entomology of the Faculty of Agronomic Sciences of the University of Abomey-Calavi located at approximately 10.0 km NW of Cotonou (Benin). The laboratory conditions were  $26 \pm 3$  °C,  $70 \pm 5\%$  RH and 12 h:12 h (L: D) photoperiod.

Mass rearing of *B. dorsalis* was conducted using the methodology described by Ekesi et al. (2007) with papaya fruits *Carica papaya* L (Caricaceae). Fifty males and fifty females of *B. dorsalis* were kept in cubical Plexiglas cages (20×20×20 cm) and provided with water in plastic container and a mixture of soybean flour and sugar in the ratio 1:3. Papaya fruit domes were punctured with entomological pin to allow easy oviposition, and offered to adult flies for 48 h. The infested papaya domes were transferred in plastic cups (8.5 cm diameter) with a complementary diet of 200 g of papaya for larval development. The cups containing the papaya fruit domes were placed each in other plastic containers (12 cm diameter and 9.5 cm depth) with a layer of sterilized sand as pupation medium. The whole incubation unit was covered with a fine mesh screen. The following days, incubated fruits were checked and dissected to allow the larvae to come out and jump into the sand. After 10-day post incubation, the sand was sieved to collect the pupae that were placed in Petri dish (5.5 cm diameter; 1.30 cm depth) then transferred in Plexiglas cages (20×20×20 cm) for adult emergence.

Fifteen-day old sexually mature adult flies were initiated to oviposition. Twenty (20) - day old fruit flies were kept into eight different cages containing 50 males and 50 females each and used in the experiments.

Punctured papaya fruits were exposed to 50 pairs of *B. dorsalis* adults for six hours. Infested fruits were thereafter exposed to 50 pairs of *F. arisanus* (four to seven-day old) for *B. dorsalis* eggs parasitization during 48 h. Emerging adults were given free access to honey droplets, water (dispensed on cotton wool) and exposed to day light for mating stimulation and oviposition. Reared on papaya fruit as larval substrate, adults of *F. arisanus* (seven- to ten- day old) were kept in four (4) different Plexiglas cages containing each 25 pairs of parasitoid and used for the experiments (Gnanvosou et al. 2016).

## Materials

Five ingredients were used in diet preparation in this study. They were papaya (*[Carica papaya L. (Brassicales: Caricaceae)]*), cassava (*[Manihot esculenta Crantz (Malpighiales: Euphorbiaceae)]*), and Moringa (*[Moringa oleifera Lam (Brassicales: Moringaceae)]*) leaves, soybean (*[Glycine max (L.) Merrill (Fabales: Fabaceae)]* flour, and hydrolysate yeast. Papaya fruits, var. solo, not initially infested, were obtained from Dantokpa, the most international market in Cotonou (Benin). Cassava leaves from eight varieties were hand-harvested early in the morning on an experimental field at IITA-Benin station, located at 12 km NW Cotonou (6°25'N; 2°19'E; 15 m ASL). These cassava varieties were 92/0067 (MR 67), 92 B/00061, 92/0057, AGRIC, I011412, I08372, I085392, and I082264 were sampled. Leaves were collected on ten plants at the eighth position starting from the first fully expanded leaf (leaf one) and counting sequentially down to leaf eight to obtain a representative sample (Bradbury and Denton 2011). Moringa leaves were sampled at the farm of the Faculty of Agronomic Sciences and the Horticulture and Genetic Units (6°25'N; 2°20'E; 28.06 m MSL). Soybean [*Glycine max (L.) Merrill (Fabales: Fabaceae)]* flour was obtained from the NGO “Terre d’amour” and used in the adult diet. The hydrolysate yeast (CAS: 100684-36-4; Affymetrix, Santa Clara, CA, USA) was obtained from the

International Institute of Tropical Agriculture (IITA-Benin) and stored in a fridge.

Moringa leaves were randomly sampled from ten seven-month-old plants at the rate of one leaf per plant, and leaves of the eight varieties of cassava samples were separately stored in transparent bags. Bags were properly labelled and conveyed to the laboratory of pharmacognosy in Porto-Novo, the historical capital of Benin, located at 30 km E Cotonou, to assess the protein contents of moringa leaves and hydrocyanic acid content of the cassava varieties. The results of protein and hydrocyanic acid contents from the various leaf samples indicated a high composition of *Moringa oleifera* and the cassava variety AGRIC in protein. The percentage of hydrocyanic acid was low in cassava samples and absent in those of *M. oleifera* (Table 1). As a result of this, we used the cassava variety AGRIC in the present study for the larval artificial diet development.

## Diet preparation and system delivery

The cassava and moringa leaves were washed thoroughly and drained on a wire mesh, in the shade. The leaves were then kept in an air-conditioned room at 21 °C for one week at the Laboratory of Agricultural Entomology of the Faculty of Agronomic Sciences of the University of Abomey-Calavi (Benin). Air-dried leaves were crushed into powder using a blender, sifted with a metal sieve to remove fibre and finally store in a tightly closed plastic container.

Papaya pulp previously removed from domes before infestation were collected in a plastic container and mashed up with the blender. It was mixed either with yeast hydrolysate, soybean flour, moringa or cassava leaves powder according to the following diets which were considered as treatments:

T1: Papaya diet (100 g).

T2: Activated yeast diet: Mixture of papaya (92 g) and activated hydrolysate yeast (8 g).

**Table 1** Protein content in moringa and cassava leaves and hydrocyanic acid content in cassava leaves

Sample code	Sample name	Water content (%)	Dry matter content (% FM)	Hydrocyanic acid content (mg/Kg)	Protein content ± sd (g/100 g of FM)	Protein content ± sd (g/100 g of DM)
1	<i>Moringa oleifera</i>	72.8	27.2	-	19.70 ± 1.20	5.36 ± 1.20
2	92/0067 (MR 67)	72.7	27.3	0.21 ± 0.10	12.30 ± 0.30	3.35 ± 0.30
3	92 B/00061	70.4	29.6	0.26 ± 0.10	11.10 ± 0.70	3.28 ± 0.70
4	92/0057	73.3	26.7	0.28 ± 0.10	12.60 ± 0.20	3.36 ± 0.20
5	AGRIC	71.2	28.8	0.05 ± 0.00	13.20 ± 0.80	3.80 ± 0.80
6	I011412	75.3	24.7	0.18 ± 0.10	8.30 ± 1.60	2.05 ± 1.60
7	I083724	76.6	23.4	0.25 ± 0.10	6.20 ± 1.30	1.45 ± 1.30
8	I085392	75.7	24.3	0.32 ± 0.10	6.70 ± 1.20	1.63 ± 1.20
9	I082264	74.9	25.1	0.27 ± 0.10	7.40 ± 1.10	1.86 ± 1.10

FM: Fresh Matter; DM: Dry Matter; sd: standard deviation

T3: Control diet: Mixture of papaya (92 g) and deactivated hydrolysate yeast (8 g)<sup>1</sup>

T4: Soybean diet: Mixture of papaya (92 g) and soybean flour (8 g).

T5: Moringa diet: Mixture of papaya (92 g) and moringa leaves powder (8 g).

T6: Cassava diet: Mixture of papaya (92 g) and cassava leaves powder (8 g).

In this study, two trials were performed using a completely randomized design. In test 1, hundred first instar larvae of *B. dorsalis* were used to infest each of the six treatments while in the test 2, hundred first instar larvae of *F. arisanus*-parasitized *B. dorsalis* were used to infest each of the six different diets. The diets were thereafter placed into incubation in the laboratory. Each diet was kept in small plastic cups (350 cm<sup>3</sup>) which was placed on a wire mesh in a larger plastic container (1200 cm<sup>3</sup>) previously provided with layer of heat-sterilized and moistened sand as pupation medium. Incubation units were covered with fine mesh screen. Ten days after incubation, the sand was sifted and pupae were collected and placed in Petri dishes. Adult parasitoids were fed on pure honey (spread over the fine white mesh of the cage) and water in wet cotton wool. Each test was replicated 6 times.

## Data collection

In test 1, parameters such as percentage of hatched eggs, percentage of pupal recovery, F<sub>1</sub> productivity, flight ability, and adult emergence rate were collected whereas in test 2, data related to the percentage of hatched eggs, percentage of pupal recovery, flight ability, and adult emergence rate were collected. Emerging adult of parasitoids were transferred all together to the bottom of a three compartments cage separated with two large holes to allow parasitoids to move from one compartment to another. The cage were covered with black plastic except at the top and set up on shelves under the light source of the rearing room. To minimize fly-back, every 10 mn, parasitoids that moved from the bottom to one of the two compartments at the top were removed and recorded. A last observation was done some hours later on the remaining parasitoids in the compartment below to identify adults that look normal but unable to fly. Flight ability was assessed using the following formula (Collins et al. 2008):

Flight ability =  $[N_{\text{pupae}} - (N_{\text{not emerged}} + N_{\text{partially emerged}} + N_{\text{deformed}} + N_{\text{non-fliers}}) / N_{\text{pupae}}] \times 100$ .

with:

$N_{\text{pupae}}$ : Number of pupae;

$N_{\text{not emerged}}$ : Unemerged puparia;

$N_{\text{partially emerged}}$ : Partially emerged puparia (part of adult stuck to the puparium);

$N_{\text{deformed}}$ : Adult emerged with deformed wings (emerged but wings deformed);

$N_{\text{non-fliers}}$ : Non flying (Adults that look normal but are not capable of flying).

A cost comparison analysis was performed to assess the profitability of the artificial diets tested. We considered the cost of ingredients needed to prepare 100 g of artificial diet.

## Statistical analyses

We conducted one-way ANOVA using diet (treatment) as the independent variable to test the significant differences in the parameter measured. Data following a multinomial distribution (percentage of egg hatch, percentage of pupal recovery, percentage of adults capable of flying) were analyzed using a Vector Generalized Linear Model (VGLM); while a Zero Inflated Poisson distribution (F<sub>1</sub> productivity) were analyzed using a Generalized Linear Model (GLM). Statistical analyses were performed using the R Statistical software (version 3.2.5, R Development Core Team 2016).

## Results

### Effects of various artificial larval diets on biological parameters of *B. dorsalis*

The highest egg hatchability was in the moringa diet. Percentage of egg hatchability higher than 30% was obtained in the cassava and control diet. The lowest hatchability was recorded in the rest of the treatment ( $P < 0.05$ ) (Table 2). However, the pupal recovery and adult emergence was higher than 80% in cassava, control diet, activated yeast, and papaya diet, which were statistically different (except pupal recovery in activated yeast). The lowest percentages were obtained in moringa and soybean diets (pupal recovery: ( $P < 0.05$ ); adult emergence:  $\chi^2 = 16.51$ ,  $df = 5$ ,  $P = 0.006$ ).

Regarding, the F<sub>1</sub> productivity of *B. dorsalis*, the highest average was obtained in the cassava diet, the lowest was in papaya diet ( $P < 0.05$ ) (Fig. 1).

### Effect of various diets on biological parameters of *Fopius arisanus*

The eggs exposed to *F. arisanus* had the highest percent of hatchability in moringa diet, which joint with the percentages with cassava diet were upper than 20%, the other diets had lower percentages ( $P < 0.05$ ). The high values of pupa recovery corresponded to control, activated yeast and moringa diets ( $P < 0.05$ ). The parasitoids emerged from each

<sup>1</sup> Deactivated hydrolysate yeast: the yeast was deactivated by keeping the activated hydrolysate yeast in an oven at 70 °C for 5 mn.

**Table 2** Effect of various larval diets on some biological parameters of *B. dorsalis* (means  $\pm$  SD) at  $26 \pm 3$  °C,  $70 \pm 5\%$  RH and 12 h: 12 h

Diets	Eggs hatchability		Pupal recovery			Adult emergence rate (%)	
	Percentage % <sup>a</sup>	Risk ratio (RR) <sup>b</sup>	Range of RR	Percentage % <sup>a</sup>	Risk ratio (RR) <sup>b</sup>		
<b>Moringa diet</b>	51.00 $\pm$ 10.10 a	2.21	[2.13; 2.30]	79.17 $\pm$ 4.36 bc	0.55	[0.52; 0.58]	58.65 $\pm$ 8.10 c
<b>Cassava diet</b>	36.17 $\pm$ 3.62 b	1.20	[1.16; 1.25]	83.50 $\pm$ 2.26 ab	0.73	[0.70; 0.78]	88.21 $\pm$ 3.52 a
<b>Control diet</b>	32.00 $\pm$ 7.90 b	1	-	87.33 $\pm$ 2.20 a	1	-	86.73 $\pm$ 2.05 a
<b>Activated yeast diet</b>	17.50 $\pm$ 1.20 c	0.45	[0.43; 0.47]	82.17 $\pm$ 2.77 b	0.67	[0.63; 0.71]	84.90 $\pm$ 3.07 ab
<b>Papaya diet</b>	15.00 $\pm$ 2.56 c	0.38	[0.36; 0.39]	87.33 $\pm$ 2.76 a	1	[0.95; 1.06]	87.99 $\pm$ 3.06 a
<b>Soybean diet</b>	15.00 $\pm$ 4.53 c	0.38	[0.36; 0.39]	76.00 $\pm$ 3.84 c	0.46	[0.44; 0.48]	72.12 $\pm$ 5.34 b

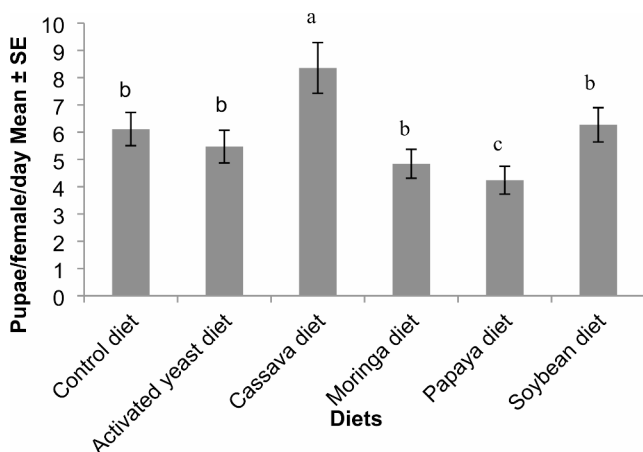
<sup>a</sup>Values within columns with different letters are significantly different (LSD test,  $\alpha=0.05$ )

<sup>b</sup>RR: Probability of *B. dorsalis* eggs to hatch and larvae to pupate in a diet compared to the probability of legs to hatch and larvae to pupate in the conventional diet consisting of hydrolysate yeast

RR < 1: Pupation or hatchability is less likely to occur in a conventional diet than in a control diet

RR = 1: There is no difference in risk compared to the control diet

RR > 1: Pupation or hatchability is more likely to occur in a conventional diet than in a control diet



**Fig. 1** Effects of various diets on F1 productivity of *B. dorsalis*. Bars followed by the same letter are not significantly different (LSD test,  $\alpha=0.05$ )

treatment had the major percentage of flight ability when the larva host was developed in papaya and cassava diet ( $\chi^2 = 2.29$ , DF = 5, P = 0.81) (Table 3).

### Cost evaluation of the artificial diets tested

The costs of ingredients used for 100 g of artificial diet were calculated based on ingredients purchased and the fraction of the mixture components (Table 4). Cassava, soybean, and moringa diets which are locally produced protein sources are 2 or 3-fold less expensive diets compared with the activated yeast and the control diets. However, the cassava diet cost is lower than the other protein-rich diets.

### Discussion

Since the high cost associated with the use of hydrolysate yeast has raised concerns, many efforts have been made in the search for alternatives (Sookar et al. 2002; Negm 2020). In the current study, we report on the potential of some locally available protein-rich including powders of moringa and cassava leaves, and soybean flour on the biological parameters of *B. dorsalis* and *F. arisanus*. The percentage of eggs hatchability, especially for *B. dorsalis* eggs subjected to parasitization (less than 30%) was lower than that reported by Ekesi and Mohamed (2011a) which should be more than 70%. This could probably be due to the egg desiccation and handling process (transfer of eggs from papaya dome to the diet). In this regard, in other experiment implementations, it will be better to search for another handling process. However, it is worth mentioning that this egg-hatching result did not affect the other parameters. In fact, these parameters were measured based on 100 first instars larvae infestation. However, the percentage of hatched eggs from moringa diet and cassava diet was higher than those from the control diet and papaya diet for both *B. dorsalis* and *F. arisanus*. The absorption of liquid from papaya by moringa and cassava leaves powder, which could also provide physical consistency and texture throughout hatching, could explain this eggs hatchability enhancement. Ekesi and Mohamed (2011a) explained that to avoid high humidity, solid-based diet should be stored at 4 °C for at least 24 h which allows the mixture to gel prior to decanting off excess water before use.



**Table 3** Effect of various host larval diets on some biological parameters of parasitoids *Fopius arisanus*

Diets	Eggs hatchability		Pupal recovery			Flight ability	
	Percentage % <sup>a</sup>	Risk ratio (RR) <sup>b</sup>	Range of RR	Percentage % <sup>a</sup>	Risk ratio (RR) <sup>b</sup>	Range of RR	(%)
<b>Moringa diet</b>	27.00 ± 2.66 a	3.15	[2.99; 3.32]	87.17 ± 3.20 ab	0.91	[0.86; 0.97]	61.67 ± 5.30 b
<b>Cassava diet</b>	21.00 ± 5.30 b	2.26	[2.15; 2.39]	80.67 ± 4.02 bc	0.56	[0.53; 0.59]	72.00 ± 8.61 a
<b>Activated yeast</b>	14.50 ± 3.71 c	1.45	[1.37; 1.53]	85.67 ± 1.96 ab	0.80	[0.76; 0.85]	61.30 ± 11.5 b
<b>Papaya diet</b>	11.67 ± 2.53 c	1.13	[1.06; 1.20]	82.00 ± 6.41 bc	0.61	[0.58; 0.65]	72.33 ± 7.63 a
<b>Soybean diet</b>	11.67 ± 2.53 c	1.13	[1.06; 1.20]	83.50 ± 5.82 b	0.68	[0.64; 0.72]	56.00 ± 9.02 b
<b>Control diet</b>	10.50 ± 3.39 c	1.00	-	88.17 ± 3.09 a	1.00	-	62.00 ± 11.3 b

<sup>a</sup>Values within columns with different letters are significantly different (LSD test,  $\alpha=0.05$ )

<sup>b</sup>RR: Probability of *F. arisanus* eggs to hatch and larvae to pupate in a diet compared to the probability of legs to hatch and larvae to pupate in the conventional diet consisting of hydrolysate yeast

RR < 1: Pupation or hatchability is less likely to occur in a conventional diet than in a control diet

RR = 1: There is no difference in risk compared to the control diet

RR > 1: Pupation or hatchability is more likely to occur in a conventional diet than in a control diet

**Table 4** Cost of each of the artificial diet tested based on unit costs of ingredients purchased

Diets	Ingredients costs <sup>a</sup> (USD)					
	Solo papaya <sup>d</sup>	Yeast <sup>e</sup>	Soybean flour <sup>f</sup>	Moringa leaves powder <sup>g</sup>	Cassava leaves powder <sup>h</sup>	Total costs
<b>Moringa diet</b>	0.17	-	-	0.056 <sup>b</sup>	-	0.27
<b>Cassava diet</b>	0.17	-	-	-	0.053 <sup>b</sup>	0.22
<b>Activated yeast</b>	0.17	0.46	-	-	-	0.63
<b>Papaya diet</b>	0.18	-	-	-	-	0.18
<b>Soybean diet</b>	0.17	-	0.0832 <sup>b</sup>	-	-	0.25
<b>Control diet</b>	0.17	0.86 <sup>c</sup>	-	-	-	1.03

<sup>a</sup> Costs were estimated from the unit price of single ingredients in CFA Franc BCEAO and then transformed to USD (exchange 1 XOF = 0.002 USD)

<sup>b</sup> Include the cost of protein sources (soybean, cassava leaves, Moringa leaves) and that of the grinding process

<sup>c</sup> Include the cost of the yeast and that of the deactivation process in the oven at 70 °C for 5 mn

<sup>d</sup> Dantokpa Market, Cotonou (500 g cost 450 XOF)

<sup>e</sup> Santa Clara, CA, USA (25 lb cost 323 699 XOF)

<sup>f</sup> NGO Terre d'amour, Abomey-Calavi, Benin (1 kg cost 200 XOF)

<sup>g</sup> Horticulture and Genetic Unit, Abomey-Calavi, Benin (630 g cost 250 XOF)

<sup>h</sup> IITA, Abomey-Calavi, Benin (800 g cost 150 XOF)

The pupal recovery obtained from both species is in line with the standard defined by Ekesi and Mohamed (2011a), which indicated that pupal recovery from the number of eggs seeded should be > 60%. It was higher than the results obtained by Lawson (2015) for a study conducted on the effect of soybean and wheat bran-based artificial diet for mass rearing of *B. dorsalis* at the Entomological Unit of the Crop Production Department (Porto-Novo/Benin). The adult emergence rate of *B. dorsalis* from the cassava diet was not significantly different from that of the control diet. Contrary to the other diets, the adult emergence rate in the moringa diet was lower than the 70% indicated by Ekesi and Mohamed (2011a). Several observations could explain this result. First of all, several pupae obtained from the moringa diet were black and small and early pupation. Secondly, early pupation noticed during the experimentation showed that moringa leaves powder has negatively affected insect

quality. The emergence rate of *F. arisanus* was not affected by the different diets used and was low in all diets (< 1%) probably due to low parasitism or larval collection process.

In this study, the lowest number of progeny obtained from papaya diet could be explained by the nutritional reserves carried over from the larval stage which provided a distinct reproductive advantage. In this study, it appeared that in spite of its relatively low protein content the cassava diet was the local protein-based diet which offers larval and adult performance close to the control diet for both species and almost all parameters considered. In contrast, regarding most parameters considered, papaya and moringa diets were lower than the control. The protein content of moringa leaves in our study is lower than the value obtained from the literature (Valdez-Solana et al. 2015). This variation in chemical analyses between our cultivar and the reference may depend on the seasonal variation, the plants' stage of

development, and the techniques employed to collect leaves samples before the experimental analysis. However, this protein content in moringa leaves in the present study was higher compared with that of cassava leaves. In addition, the protein content in moringa leaves and soybean was higher than that of cassava but did not offer the best performance on the mass-rearing parameters of *B. dorsalis* and *F. arisanus*. The difference observed between activated yeast diet and the control diet on *B. dorsalis*, for parameters such as egg hatch and pupal recovery could be explained by the better digestibility of deactivated yeast compare to the activated one. However little is known in the literature about the difference between the two yeast products and therefore, needs further investigation.

In the present study, the cassava diet outperformed other diets. That may be attributed to its composition in amino acids and protein which is highly digestible. For example, FAO/WHO (1973) reported that the essential amino acid content of cassava leaves is higher than soybean protein. This highlights the importance of evaluating the nutritional composition of each product. Moreover, the various locally produced protein sources tested in our study are cost-effective compared to the yeast. The cassava diet cost in particular is lower than the other protein sources. This can be explained by the use of cassava leaves more often for direct consumption by humans or livestock than for sale. In addition, leaves are rather considered residues and they do not compete with the roots, which are the main commercial product of cassava. This observation coupled with the significant results of cassava for biological parameters on the two species can lead to the recommendation of cassava diet as a substitute for the expensive yeasts. The composition of the insect diet in essential amino acids, proteins, vitamins or cholesterol is vital for larval development, pupal recovery, pupal weight, adult emergence, and flight ability. Chapman et al. (2013) highlighted that nutrient availability during larval feeding provides pupa and adults with the needed resources to survive. This suggests that diets provided to larvae during mass-rearing must be potentially rich in terms of nutritional composition to complete the life cycle. In the present study, even though the cassava diet showed promising results, the adaptation process of fruit fly strains needs to be further investigated and scaled up. Thus, assessing the efficiency of these different diets on fruit flies and parasitoids over several generations is of utmost importance as several generations are required for the insects to adapt to artificial diets (Economopoulos 1992; Ekesi and Mohamed 2011a).

## Conclusion

In summary, the current study investigated the efficiency of some local protein-rich diets (moringa leaves powder, cassava leaves powder, soybean flour) on *F. arisanus* and *B. dorsalis* performance, compared with the deactivated and activated hydrolysate yeast. It showed that cassava diet was the local protein-rich diet that offered better performance on the life history parameters of *F. arisanus* and *B. dorsalis* close to the control diet. Then, cassava diet which is also cost-effective can conclusively be used to replace the imported hydrolysed yeast. These results are great of importance for the establishment of efficient control of fruit flies using locally available larval diets which are vital for parasitoids survival and performance.

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## Declarations

**Conflict of interest** The authors declare no conflict of interest.

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