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Rearing of palmetto weevil, *Rhynchophorus cruentatus* fabricius (Coleoptera: Curculionidae) under laboratory conditions

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Abstract
Palmetto weevil (*Rhynchophorus cruentatus*) (Coleoptera: Curculionidae) is considered North America's largest weevil (Weissling and Giblin-Davis, 1997) [21]. It is a tissue borer to different palm trees, especially sabal palms (*Sabal palmetto*). It is a resident of Florida but has been found in south Texas to the west till South Carolina to the north (Thomas, 2010) [20].
Like other weevils of the genus *Rhynchophorus*, it has a cryptic habitat; many generations can live inside the palm trunk for the entire life cycle. Larvae are the most dangerous stage; they penetrate deep into the palm trunk, feeding on soft tissues and causing total loss of the palms. Control strategies for this weevil mainly depend on using chemical insecticides that can be used for curative treatments, but persistence is necessary (Weissling and Giblin-Davis, 1997) [21]. New eco-friendly control methods should be investigated for environmental preservation as substitutes for chemical insecticides (Isman, 2000) [15]. Testing new control methods against the palmetto weevil in the laboratory requires the availability of different insect stages. Therefore, this study was designed to improve an efficient, easy, and cheap rearing technique to establish a colony of palmetto weevils in the laboratory, thus providing enough different developmental life stages for further studies and laboratory bioassays.
Adult weevils were collected from the field using food-bait pheromone traps. Seven traps were installed in Brunswick, GA, USA. Pheromone traps provided us with adult individuals, males, and females, which were needed as initial samples for establishing the palmetto weevil colony in the laboratory. The cumulative numbers of collected weevils during a trapping period from October (2019) to May (2020) are 505, with a 1:1.8 sex ratio between males and females. The rearing process was carried out in a controlled room at the Entomology Department, University of Georgia, USA. The room temperature was maintained at 26 ± 2ºC, and humidity was maintained at 60-80% RH. The photoperiod was approximately 16:8 L: D. Field Collected adults were provided with 20 gm apple slices for feeding and as egg-laying substrate. Eggs were collected daily, and hatched larvae were provided individually with apple slices for feeding; however late larval stages were provided with sugarcane stems to complete metamorphosis. Sugarcane fibers were essential for larvae to construct cocoons and enter pupation. The duration of each developmental life stage was recorded. Eggs required 3.71 ± 1.137 days to hatch. Ten larval instars were observed during the life cycle as the larval stage required 79.21 ± 12.994 days to enter pupation. Newly hatched adults emerged from the cocoons after 25.63 ± 6.47 days. The mean generation time was estimated as 110.3 ± 21.3 days. The palmetto weevil colony was established and maintained successfully under laboratory conditions using an apple-sugarcane diet.

Keywords: laboratory rearing, *Rhynchophorus Cruentatus*, palmetto weevil, palm trees, pheromone trapping, and apple-sugarcane diet

Introduction
The genus *Rhynchophorus* is considered to have ten described species of weevils that infest different types of palm trees. Palmetto weevil (*Rhynchophorus cruentatus*) is North America's largest weevil (Weissling and Giblin-Davis, 1997) [21]. It is a resident of Florida but has been found as far as south Texas to the west and South Carolina to the north (Thomas, 2010) [20]. Fossils suggested that the palmetto weevil may have been present in Florida since one million years ago (Shane Tedder *et al.*, 2012) [18]. It is a concealed tissue borer to different palm trees, including cabbage palms (*Sabal palmetto*), Canary Island date palms (*Phoenix canariensis*), date palm (*P. dactylifera*), Latan palms (*Latania sp.*), royal palms (*Roystonea sp.*), *Washingtonia sp.*, Bismarck palms (*Bismarckia nobilis*), and coconut palm (*Cocos nucifera* Linnaeus) (Weissling and Giblin-Davis, 1997) [21]. Palmetto weevil is holometabolous with a complete life cycle. Adult's length ranges from (25mm to 45mm). Female weevils deposit up to 800 eggs in the holes and wounds in the palm trunk (Thomas, 2010) [20]. Like other weevils of the genus *Rhynchophorus*, larvae are the most dangerous stage; they penetrate deep into the crown and the stem using their long rostrums, making tunnels, feeding on soft tissues, and disrupting the palm's vascular system, causing trunk weakness and total loss of the palms. Late larval instars used palm tissue fibers to construct cocoons. All developmental stages remain inside the palm trees for generations without apparent symptoms. The infestations cannot be detected in the early stages leading to difficulties in treatment. Recently, there has been an observed increase in palms killed by palmetto weevil (*R. cruentatus*), and the infestation is distributed to reach palms at local palm nurseries and public parks in South Georgia and North Florida. Palmetto weevils were responsible for destroying two hectares of canary island palms in Florida, as only 24 larvae are enough to kill one palm tree (Hunsberger

et al., 2000). Also, it is economic importance came from it is predicted as a potential vector of red ring nematode (*Bursaphelenchus cocophilus*) that causes palm red ring disease (Giblin-Davis and Howard, 1988) [10].

Controlling *R. cruentatus* depended mainly on chemical insecticides (Giblin-Davis and Howard, 1988) [10]. Previous laboratory investigations were conducted to detect the impact of different commercially available pesticides in controlling palmetto weevil adults (Giblin-Davis and Howard, 1989) [11]. The tested insecticides were: Lindane, chlorpyrifos, propoxur, dimethoate, and methomyl. The tested insecticides effectively killed the weevils compared to the control, except methomyl (Giblin-Davis and Howard, 1989) [11]. Recently, chemical control has become undesirable since chemicals lead to harmful effects on the environment and the evolution of pest resistance against insecticides (Isman, 2006) [14] and because of the cryptic habit of insects. Thus, safer and more effective control approaches should be explored against this economically important pest. Testing new control methods against the palmetto weevil in the laboratory requires the availability of different insect developmental stages, so improving laboratory rearing techniques is required. Previous studies reported different rearing techniques of *Rhynchophorus sp*., especially *R. ferrugineus*, in the laboratory (Shahina *et al*., 2009; Al-Ayedh, 2011; Sharaby and Al-Dhafar, 2013; Norzainih *et al*., 2015; El-Zoghby and Abdel-Hameid, 2018) [17, 2, 19, 16, 7].

In the case *of R. cruentatus,* much fewer rearing trials were conducted to establish a colony in the laboratory (Giblin-Davis *et al.*, 1989; Weissling and Giblin-Davis, 1994, 1995). Giblin-Davis *et al.* (1989) [9, 23] successfully reared *R. cruentatus* under laboratory conditions using a pineapple-sugarcane diet, but it was noted that using a pineapple has some restrictions. Pineapple is not preferred because it decomposed to liquefication which is believed to be harmful to the neonate larvae. Pineapple was reported to be a difficult egg-laying substrate because it is hard to dissect it to remove eggs (Giblin-Davis *et al*., 1989) [9]. According to Weissling and Giblin-Davis (1994) [23], apple slices are supposed to be easier to dissect than pineapple, thus facilitating egg removal. However, weevils that have been reared using only apple slices reached the 4th larval instar and failed to complete the life cycle due to the absence of fibers needed to construct cocoons (Shahina *et al.*, 2009) [17]. It is observed that sugarcane stems are a common component of various rearing diets because their fibers are essential for constructing cocoons to complete the life cycle in the laboratory. Sugarcane fibers work as a substitute for palm tissue fibers. On the contrary, high prices and less availability in different seasons in Georgia, USA, represent limitations to using sugarcane fibers as a single rearing diet.

Therefore, this study was designed to improve an efficient, easy, and cheap rearing technique to establish a colony of palmetto weevils under laboratory conditions, thus, providing enough different developmental life stages needed for further studies and laboratory bioassays. A combination of the apple-sugarcane diet was evaluated in this research as a rearing technique for *R. cruentatus.* This apple-sugarcane combination is supposed to overcome the restriction of using each diet separately.

Materials and Methods

Insect sampling

Food bait pheromone traps were used to attract and collect adult populations of the palmetto weevils from infested palms located at 31°9'51.85" N latitude 81°30'20.16" W longitude in Brunswick, GA, USA. A total of 7 traps were installed in the forested area, specifically the maritime hammock habitat, where no pesticides were applied for a trapping period from October (2019) till May (2020).

Pheromone traps were designed according to (Hanounik *et al.*, 2000). Each trap was designed using a 5 L plastic bucket (Fig. 1a) with 4 holes in the bucket's lid and 4 lateral rectangular windows around the side walls. The outer surface of the bucket was covered by dark, rough cloth (sackcloth) to help the weevils climb it and enter rather than falling from the smooth surface of the bucket; also, the dark color is more effective in catching weevils (Al-Saoud, 2018) [3].

Components of Pheromone traps; Each trap bucket carried the following materials: 1) Pheromone lure: a Pack of 400 mg cruentol® (Synthetic *R. cruentatus* male aggregation pheromone) with the active ingredient (5-Methyl-4-octanol) (Weissling *et al.*, 1994), imported from Chem Tica Internacional SA, Heredia, Costa Rica (Fig. 1b), 2) A 20 ml bottle of kairomone (Ethyl acetate with 98% purity), 3) Food substrate (350 gm Pineapple), 4) A teaspoonful of yeast, and 5) 2 L of water, creating a water level inside the trap below the windows. The underside of the lid had a small knob to which a wire was fixed to hold the pheromone lure and kairomone bottle dispensers (Fig. 1b). The trap was insecticide-free pheromone traps to collect alive weevils needed for the laboratory assay. Kairomone (ethyl acetate) was added to pheromone traps to increase the efficacy of collecting weevils (El-Shafie and Faleiro, 2020). Traps with food substrate were proved to attract more weevils and keep these captured weevils alive (Al-Saoud, 2018) [3].

Placement and serving of pheromone traps: Seven traps were buried in the soil up to the lateral holes (Fig. 1c). Checks on the trap were carried out twice a week. The pheromone pack was replaced by a new one almost monthly, the kairomone bottle was refilled when needed, and the food substrate (pineapples) changed weekly. Water in the traps was replenished as needed to maintain sufficient moisture. Captured weevils were collected from each trap twice a week (Fig. 1d), and identification was made according to the morphological characteristics of *R. cruentatus* (Giblin-Davis *et al.*, 2013) [8].

Captured weevils were moved to the laboratory, where they were counted and sexed based on the shape and length of the rostrum. The weevils were maintained under laboratory conditions (26 ± 2 °C, 65±5% RH, and photoperiod from 18:6 L:D) in plastic jars as initial individuals to establish a palmetto weevil colony.

Rearing experiment

The rearing technique is based on using the apple-sugarcane diet, a modified method from the previous work by Giblin-Davis *et al.* (1989) [9], as pineapples were replaced with apples which are cheaper, easier to dissect, and available in local stores.

Rearing room

The rearing process was carried out in a controlled rearing room at The Entomology Department, University of Georgia, USA. The room (Fig. 2a) was maintained at 26 ±

2ºC and 60-80% RH. The photoperiod was approximately 16:8 L: D.

Equipment and materials required for the rearing process include small, medium-sized (Fig. 2b), and large plastic jars, Petri dishes, balance, wetted filter paper (Fig. 2d), wetted cotton, paintbrush (Fig. 2f), Apple slices (Fig. 2c & e), and Sugarcane stems (Fig. 2g & h).

Insect maintenance

Field collected adults provide the initial samples for rearing to establish the palmetto weevil's colony. They were counted and sexed, and males were separated from females. Adults are maintained at 26 ± 2 ºC and 60-80% RH with 16:8 L: D. and provided with apple slices (20 gm) as food and egg-laying substrate and wetted cotton for keeping them moist**.** Food was replaced every three days.

Breeding and handling developmental stages

A male and 2 females were put in medium-sized covered vented plastic containers to allow the breeding, and this jar contained an apple slice (20 gm) for feeding and egg-laying. Wetted cotton was added for moisture. Eggs were collected daily.

Egg: Apple slices, loaded with eggs, were removed from adult jars and replaced with new apple slices. Collected eggs were transferred and put in a petri dish with a wetted filter paper (Fig. 2d) and kept at 26 ± 2 ºC and 60-80% RH till hatching. Eggs were checked every day to harvest hatched larvae.

Larvae: Newly hatched larvae were harvested, transferred to separated plastic jars, and provided with apple slices as a food source and wetted cotton for moisture **.**Newly hatched larvae were handled using a paintbrush (Fig. 2f). Food was replaced every three days. Different parameters of larval development were recorded weekly, including weight, length, head width, and head length, to detect different larval instars.

Pre-pupa: Late larval instars were transferred to new plastic jars and provided with apple slices in addition to sugarcane stems of suitable size (Fig. 2 h). (Size 5×2 cm) as a food source and source of fibers essential to constructing cocoons and completing the life cycle.

Pupa: After cocoon construction, late larval stages stopped feeding and moving and entered the pupation period. Collected pupae were transferred to new plastic jars and kept at 26 ± 2 ºC and 60-80% RH until adult emergence.

Adult emergence: By the end of the pupation period, fully grown adults began to emerge from cocoons. Emerged adults were sexed and transferred to separate new plastic jars.

Statistical analysis

The sum of the numbers of collected weevils was gathered cumulatively throughout the trapping period from October (2019) till May (2020). The differences in the sex ratios for captured males and females were statistically analyzed. The duration of each developmental life stage was recorded. Mean ± SD time for each life stage was calculated using the descriptive statistics function in Microsoft Excel (2019). Mean ± SD morphological parameters of different larval instars were detected. Mean ± SD generation time was also calculated.

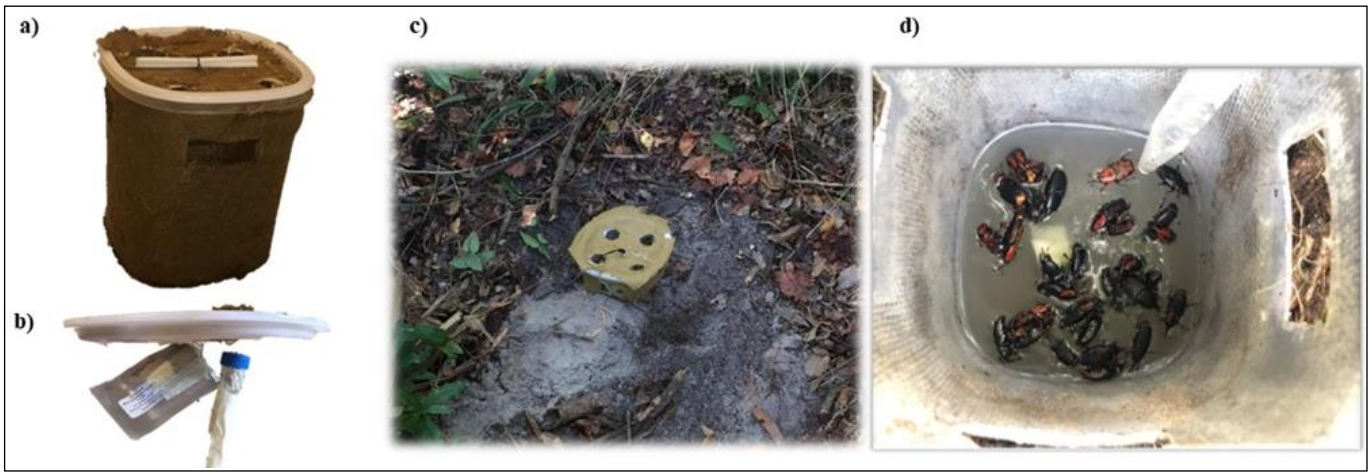


Fig 1: Pheromone traps: (a Plastic trap bucket covered with sackcloth, (b the lid of the trap bucket holding the Pheromone pack and the kairomone bottle, c) Placement of the pheromone trap between the palm trees, and d) Collected palmetto weevils from the pheromone trap.

Fig 2: a) Controlled rearing room, **b)** Plastic jars containing adults, **c)** Plastic jars containing apple slices and sugar cane stems, **d)** Collected eggs on wet filter paper, **e)** Apple slices loaded with larvae, **f)** A paintbrush, carrying 1st larval instar, **g)** Sugarcane stems, and **h)** Sugarcane stems loaded with larvae.

Results

Numbers of captured weevils and determination of the sex ratio

A total of 7 food-bait pheromone traps were installed to capture adult weevils in Brunswick, GA, USA. Pheromone trapping was a successful method to capture adult palmetto weevil during a trapping period from October (2019) till May (2020).

Results showed that the cumulative number of captured weevils using the pheromone traps during the trapping period from October (2019) till May (2020) was (505) weevils, (324) of them were females, and (181) were males (table 1). According to figure (3), females represent (64%) of the total weevil numbers, and (36%) represent the percentage of captured males.

Sex ratio between males and females of captured Palmetto weevil

As shown in table **(1)**, the number of captured females is significantly more than the number of captured males **(P < 0.05),** with a sex ratio equal to **(1 male: 1.8 female)**. This proves that female weevils responded to the aggregation pheromone more than males.

Table 1: Variations between the captured number of males and females Palmetto weevils using pheromone traps.

Sex	No. of weevils	Sig.	Sex ratio (male/female)
Male	181[b]	0.037144	1: 1.8
Female	324[a]		
Total	505		
Different letters refer significant differences (t-test, P < 0.05)			

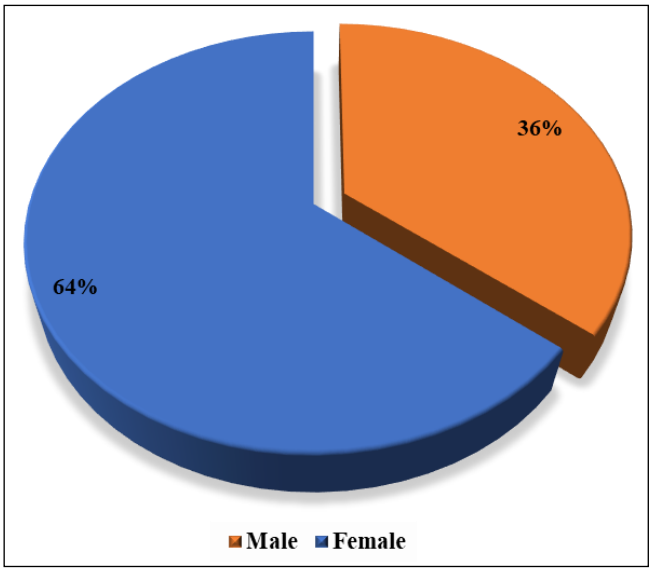


Fig 3: Variations between the captured number of males and females Palmetto weevils using pheromone traps.

Life cycle and developmental stages of palmetto weevil

Palmetto Weevils (*R. cruentatus*) were successfully reared using an apple-sugarcane diet under laboratory conditions. The time length of each developmental stage in the life cycle was recorded. The mean time of each period was calculated to determine the mean generation time of the palmetto weevil.

Pre-oviposition period

The pre-oviposition period is the period between female emergence and laying eggs. As shown in table (2), females take a period ranging from 1 to 3 days, with 1.76 ± 0.744 days as an average time needed before laying eggs (Fig. 4a).

Egg hatching

The egg hatching period is the time needed for the eggs to hatch. Table (2) showed that the egg hatching period ranged from 2 to 6 days, with 3.71 ± 1.137 days as an average time needed for eggs to hatch into larval stages. As shown in figure (4b), eggs are small, elongated, and creamy white.

Larval stage

Larvae are creamy white and legless, with a well-developed head capsule. Larvae are the most destructive stage, having biting chewing mouth parts that help to penetrate through soft palm tissue (fig. 4c). The larval stage period ranges from 50 to 101 days, with 79.21 ± 12.994 days as an average time needed for the larva to transform to pupa (table 2). Different larval instars were observed (fig. 4d) as the larva could molt up to 10 times**.** Head width and length were measured weekly, and means were illustrated in table **(3)** to differentiate between different larval instars.

Pupal stage

Late larvae in the pre-pupal period stopped feeding and started to construct the cocoon from sugarcane fibers. They stopped moving and entered pupation (fig. 4e, f &g). The Pupal stage period ranged from 14 to 36 days, with 25.63 ± 6.470 days (table 2) as the average time required for a new adult to emerge from the cocoon.

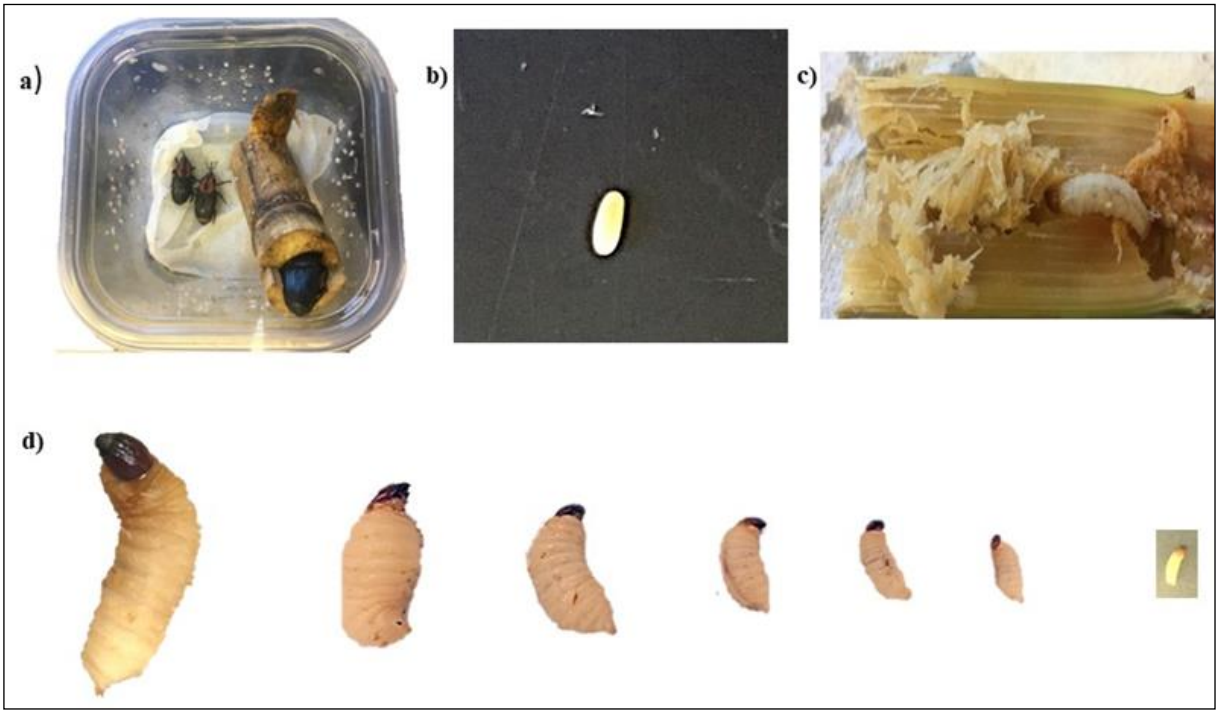
Table 2: shows the mean duration of different developmental stages during the *R. cruentatus* life cycle.

Developmental Stage	n	Duration (days)		
		Mean	SD	Range
Pre-oviposition	50	1.76	0.744	1-3
Egg hatching	49	3.71	1.137	2-6
Larva	47	79.21	12.994	50-101
Pupa	43	25.63	6.470	14-36

*n refers to the number of samples

Table 3: shows morphological parameters of different larval instars of *R. cruentatus.*

Larval instar	Head length (mm)	Head width (mm)	Length (mm)	Weight (gm)
	Mean ± SD			
1	2.046 ± 0.315	0.874 ± 0.217	11.06 ± 2.237	0.0322 ± 0.007
2	3.052 ± 0.347	1.76 ± 0.153	15.848 ± 1.174	0.0675 ± 0.013
3	4.064 ± 0.578	2.62 ± 0.23	18.914 ± 0.838	0.126 ± 0.024
4	4.864 ± 0.338	3.42 ± 0.326	21.776 ± 0.819	0.1851 ± 0.020
5	5.862 ± 0.447	4.442 ± 0.314	24.496 ± 0.893	0.2525 ± 0.031
6	6.784 ± 0.425	5.214 ± 0.135	26.948 ± 1.038	0.4591 ± 0.164
7	7.93 ± 0.357	5.724 ± 0.154	33.492 ± 2.833	1.0871 ± 0.217
8	8.902 ± 0.307	6.24 ± 0.163	37.56 ± 0.620	1.7622 ± 0.029
9	10.028 ± 0.445	6.732 ± 0.185	38.828 ± 0.449	1.9237 ± 0.076
10	11.01 ± 0.385	7.316 ± 0.188	42.7 ± 2.280	2.1887 ± 0.133



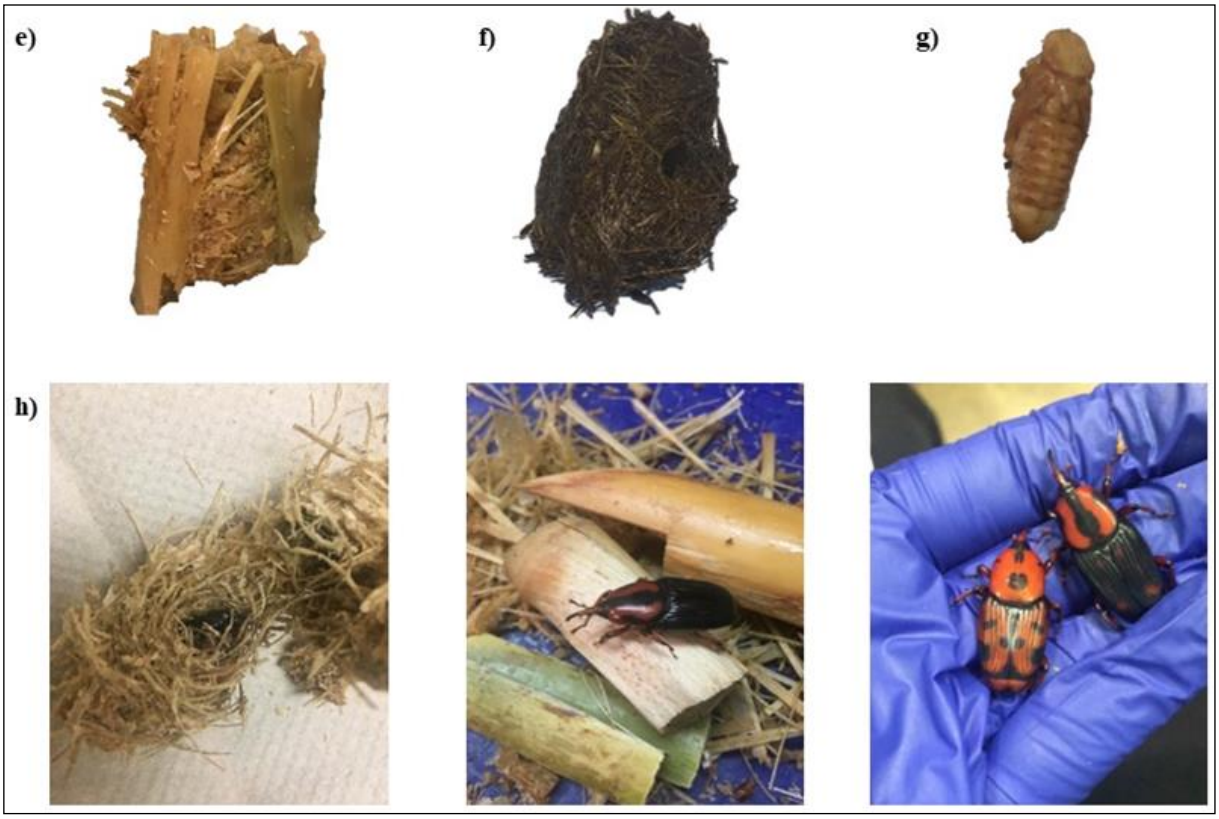


Fig 4: Different developmental stages during *R. cruentatus* life cycle, a) Palmetto weevil adults (1 male & 2 females) prepared for breeding, b) Egg, c) Larva boring through sugarcane stem, d) Different larval instars during Palmetto weevil life cycle, e) Cocoon, made of sugarcane fibers, f) Pupa with a cocoon, g) Pupa without a cocoon, and h) Adult emergence

The mean generation time

The mean generation time to complete one generation of palmetto weevil under laboratory conditions using an apple-sugarcane diet was 110.3 ± 21.3 days, approximately 16 weeks. Mean generation time ranges from 67 to 146 days (table 4).

Table 4: shows the mean generation time of *R. cruentatus*.

Mean (days)	SD (days)	Range (days)
110.3	21.3	67-146

Adult emergence

At the end of the pupation period, fully grown adults started to emerge from the cocoons. The average life span of males and females was illustrated in table **(5)** as females' average life span is about (26.599) weeks, in contrast with 28.626 weeks for males. Males significantly live more than females (P=0.003218). Regardless of sex, the life span of an adult weevil ranges from 131 days to 285 days.

Adult weevils have different color patterns, ranging from all black to reddish brown with black spots. The prothorax is the first thoracic segment of the body of the adult weevil. It is cone-shaped and suddenly narrowed from its anterior part to give the shape of shoulders. Pronotum has black marks scattered on its surface. It articulates anteriorly with the head and posteriorly with the second thoracic segment (mesothorax). The unidentate mandibles from mouth parts are found at the end of a long prominent curved snout (rostrum), representing one-third of the entire body length.

The shape of the snout distinguishes male and female weevils. The male snout appeared rough and slender, with tiny tubercles at the distal part. Unlike red palm weevil males, dorsal rostral hairs are absent in palmetto weevil males. Female snouts are smooth, lack tubercles, and more curved, and are longer than males. A pair of elytra are carried on the second thoracic segment (mesothorax). The elytron is the fore wing of adult palm weevils; it is hard, fully sclerotized, and red with furrows on its dorsal side. Elytra cover the membranous hind wing forming a protective shield during rest.

Table 5: shows the mean life span of *R. cruentatus* adults.

	n	Mean (Weeks)	SD	Range
Female	50	26.599[b]	0.4273	19.9 - 40.7
Male	50	28.626[a]	0.5197	18.7 - 40.4
Total	100	27.613	1.1891	18.7 – 40.7

Different letters refer to significant differences (t-test, P < 0.01).

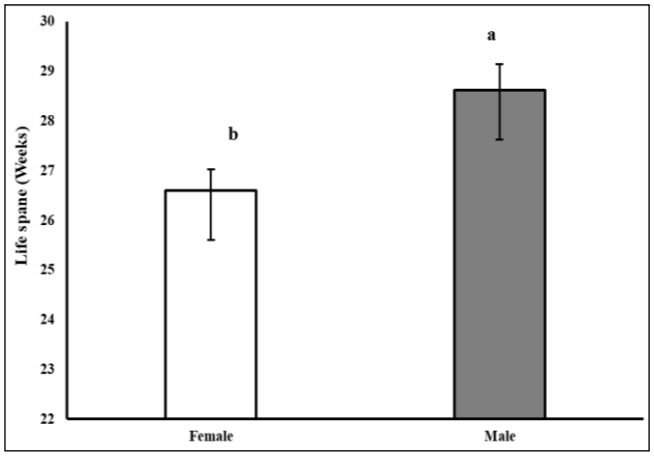


Fig 5: Variations between the mean life span between males and females Palmetto weevils. Columns represent the means (±SD). Different letters refer significant differences from the control group (t-test, P < 0.01)

Discussion

In the current study, Palmetto weevils (*R. cruentatus*) were successfully reared on an apple-sugarcane diet under laboratory conditions. A palmetto weevil colony was established, and enough different developmental life stages required for further studies and laboratory bioassays were available.

The rearing technique in this study is a modified technique of the previous work of Giblin-Davis *et al.* (1989) [9], but with replacing pineapples with apples because using pineapples has some restrictions. Pineapple is decomposed to liquefication which is harmful to the neonate larvae. Pineapple was a difficult egg-laying substrate because it is hard to dissect it to remove eggs (Weissling and Giblin-Davis, 1994) [23]. Apples were selected as the main food source and egg-laying substrate because they are cheap, easy to handle, and available around the entire year in Georgia, USA, local stores. According to Weissling and Giblin-Davis (1994) [23], apple slices are supposed to be easier to dissect than pineapple, thus facilitating egg removal. The fecundity of palmetto weevils was studied using apples as an egg-laying substrate (Weissling and Giblin-Davis, 1994) [23]. An essential outcome of this study was that adult weevils had 8 times more fecundity when using apples instead of pineapple. On the other hand, weevils that have been reared using only apple slices reached the 4th larval instar and failed to complete the life cycle due to the absence of fibers needed to construct cocoons (Shahina *et al.*, 2009) [17].

Previous literature reported that sugarcane stems are a common component of various rearing diets because their fibers are essential for the late larval instars to construct cocoons and complete the life cycle in the laboratory (Shahina *et al.*, 2009; Al-Ayedh, 2011; Sharaby and Al-Dhafar, 2013; Norzainih *et al.*, 2015; El-Zoghby and Abdel-Hameid, 2018) [17, 2, 19, 16, 7]. Sugarcane fibers work as a substitute for palm tissue fibers. Despite the efficacy of sugarcane in rearing techniques, high prices and less availability in different seasons in Georgia, USA, represent limitations to using sugarcane fibers as a single rearing diet. Therefore, this study was designed to improve an efficient, easy, and cheap rearing technique to establish a colony of palmetto weevils under laboratory conditions. Thus, A combination of the apple-sugarcane diet was evaluated in this research as a rearing technique for *R. cruentatus.* This apple-sugarcane combination succussed in overcoming the restriction of using each diet separately.

The pheromone trapping technique is an important component of integrated pest management programs; it helps capture adult insects from the field and decreases their populations (Dhouibi *et al.*, 2017). This research used pheromone traps to capture alive adult weevils from the field to use them as initial samples for colony establishment. The cumulative numbers of collected weevils during a trapping period from October (2019) to May (2020) are 505, with a 1:1.8 sex ratio between males and females. Similar results were reported by Shane Tedder *et al.* (2012) [18], as they found a significant difference between collected numbers of males and females, with more females attracted to the traps. Our results correspond with Abbas *et al.* (2006), who found that *R. ferrugineus* females were more likely to respond to the aggregation pheromone than males (Sex ratio = 1 male: 1.5 Female). El-Bokl *et al.* (2015) reached the same conclusion with a 1 male: 2 females sex ratio when using the pheromone trapping technique in (Damietta, Egypt) date palm plantations.

Field collected adults were used as initial samples for establishing the palmetto weevil colony under laboratory conditions (26 ± 2 °C, 65±5% RH, and photoperiod from 18:6 L:D). The duration of each developmental stage during the insect life cycle was recorded. Our results showed that the mean generation time was approximately 16 weeks (110.3 ± 21.3 days) with a range of (67-146 days). Giblin-Davis and Howard (1988) [10] reported that one generation required less than 84 days to complete in nature. Rearing palmetto weevil in the laboratory at 23 ºC using sugarcane stems showed an average generation time of more than 212 days. (Giblin-Davis *et al.*, 1989). Thomas (2010) [20] recorded that (45-180 days) were required to complete one generation of *R. cruentatus*. It is documented that nutrition, temperature, and humidity are key factors that affect generation time (Giblin-Davis *et al.*, 2013); this may explain variation in the generation time within the same species.

Data obtained from this result revealed that the pre-oviposition period was 1.76 ± 0.744 days with a range from 1 to 3 days, and eggs needed from 2-6 days to hatch. Previous observations in nature recorded that 64 h was the needed time for hatching (Giblin-Davis *et al.*, 1989). In the laboratory, each female deposited up to 26 ± 15 eggs, 69 ± 17 hours was recorded as an egg hatching period. The average weight of the last larval instar was (3.5-5) gm (Giblin-Davis *et al.*, 1989). Thomas (2010) [20] recorded that the egg-hating period was about 3-4 days. The larval stage has the longest period between immature stages with up to 10 instars. Ten larval instars were observed during the life cycle as the larval stage required 79.21 ± 12.994 days to enter pupation with a range of (50-101) days. Compatible with our results, active larval duration ranged from 25 to 105, but nine to twenty larval instars were observed (Thomas, 2010) [20]. The larval stage duration in *R. ferrugineus* larvae ranged from 25 to 105 days till pupation, which required 11 to 50 days (Giblin-Davis *et al.*, 2013). Our records detected that newly hatched adults emerged from the cocoons after 25.63 ± 6.47 days of pupation and ranged from (14-36). Previous rearing experiments showed that pupation could take up to two weeks (Giblin-Davis *et al.*, 1989) [9]. Thomas (2010) [20] results observed that 2 to 17 days were required to prepare larvae for pupation and the pupation period lasted from 8-50 days. Another study mentioned that the pupation period ranged from 33 ± 13.3 to 46 ± 2.9 days (Weissling and Giblin-Davis, 1995) [22].

In this study, the average life span was illustrated; females' average life span is about (26.599) weeks, in contrast with 28.626 weeks for males. Males significantly live more than females. Regardless of sex, the life span of an adult weevil ranges from 131 days to 285 days. Similar to our findings, Giblin-Davis and Howard (1988) [10] found that adult male lifespan was longer than female lifespan as males lived for 87 ± 41 days and females survived 74 ± 16 days. A similar observation noted that the male life span was longer than the female's (Giblin-Davis *et al.*, 1989). Adult weevils could live more than 100 days (Thomas, 2010) [20].

Thus, data obtained from this project will help to increase our knowledge about the biology of the palmetto weevil. Also, further investigations could be conducted using individuals produced from establishing this colony to evaluate new methods to control this economic pest. This

project is the first to rear palmetto weevils (*R. cruentatus*) under laboratory conditions using an apple-sugarcane diet.

Conclusion

In conclusion, The Palmetto Weevils, *Rhynchophorus cruentatus*, were successfully reared using an apple-sugarcane diet under laboratory conditions. Apples are cheap, easy to handle, and available in Georgia, USA local stores. Sugarcane fibers are essential to constructing cocoons. Life parameters were recorded, including pre-oviposition, oviposition and larval & pupal periods, adult male and female lifespans, and mean generation time. The mean generation time is approximately **16** weeks with a range of (67-146 days). The larval stage has the longest period between immature stages with up to 10 instars. This project is the first to rear palmetto weevils (*R. cruentatus*) under laboratory conditions using an apple-sugarcane diet. Data obtained from this project will help to increase our knowledge about the biology of this weevil. Also, further investigations could be conducted using individuals produced from establishing this colony to evaluate new methods to control this economic pest.

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