S. Pac. J. Nat. Sci., (2002), Vol. 20, pp. 22 - 24
Section A: Short Communication

Sex Ratio in Hawaiian Papaya (*Carica papaya* L.) Variety 'Solo'

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Abstract

Fruits from hermaphrodite Hawaiian varieties are of medium size and preferred by consumers all over the world. Seeds obtained from the fruits of selfed hermaphrodite plants produce almost two third hermaphrodite plants and one-third pistillate plants. These plants can not be identified before the onset of flowering. However, there are conflicting reports in scientific literature on sex ratios obtained from selfed seeds from fruits of hermaphrodite plants. Sex ratios of hermaphrodite, pistillate and staminate plants were determined using seeds from selfed hermaphrodite fruits of a Hawaiian papaya (Carica papaya) variety 'solo'. Bagging flowers on hermaphrodite plants during flowering season to avoid contamination from foreign pollen produced self-seeds. Seeds from fruits of many bagged hermaphrodite flowers were pooled together and grown in large plots. A total of 2633 plants were grown and evaluated for their specific sex at the onset of flowering. Of these 1838 were counted as hermaphrodite, 783 pistillate and only 12 staminate plants. This gave a ratio of 2.34 hermaphrodite plants : 1: pistillate plants ignoring 12 staminate plants. This obtained ratio is in disagreement with earlier reported ratio of 2 hermaphrodite plants : 1 pistillate : 1 non-viable zygotes. However, no viable male progeny was reported from hermaphrodite selfed seeds in earlier studies. One of the explanations for the observed changed ratio could be biased seed selection from the seed lot used for planting.

1 General Introduction

The medium sized Hawaiian pawpaw is the preferred one for consumption and these fruits are borne on the hermaphrodite (bears both male and female flowers) plants. The female plant also bears fruits but they are not as good. When seeds are sown only about 2/3 plants are hemorphrodite and 1/3 are pistillate (female). These plants can not be differentiated until they start to flower and the problem then is contamination of flowers on the hermaphrodite plants with the pollen of female plants. To avoid contamination flowers of the hermaphrodite plants must be covered to allow self pollination to occur. There is some indication that the actual ratio of hemorphrodite to pistillate plants can change due to some environmental factors such as temperature and thus there are some conflicting reports on sex ratios. This study was carried out to determine the sex ratio in these plants in Fiji under our local weather conditions.

Our results showed that the ratios obtained were not strictly in a 2:1 ratio, thus suggesting some possible influence causing the variation. One simple possibility could be that the seed selection was biased since we may have chosen not to choose seeds that looked less healthy. However, there could be more complex reasons that need to investigated.

2 Introduction

Papaya *(Carica papaya)* is the only economic species of the genus *Carica*, which has 21 species and is the largest genus in the family Caricaceae.. Papaya plants are either hermaphrodite (flowers with both sexes), pistillate (flowers with female sex only) or staminate (flowers with male sex only). Staminate plants flower earlier and produce male flowers in large numbers on branched stalks (Singh 1990). They are funnel-shaped about 2.5cm long, five white petals joined towards the base and ten stamens near the opening of the tube (Singh 1990, Hastie 1994). Though normally male (staminate) flowers cannot produce fruit, some older plants may produce bisexual flowers during cooler months, which form small-elongated fruits (Hastie 1994). Female (pistillate) flowers are relatively large, borne singly or in small clusters in the axis of leaves. They have a functional pistil but no stamens, five fleshy white petals, five sessile fan-shaped stigmas and a large ovoid-shaped superior ovary (IBPGR Report 1986; Popenoe 1974). Cross-pollination by wind and / or insects is necessary for fruit development in pistillate flowers. Gynodioecious lines produce female (pistillate) and hermaphrodite (bisexual) flowers (Manshardt and Drew 1998). Hermaphrodite lines produce bisexual flowers that have both male and female parts. They are larger than staminate flowers with petals joined at base, pollen-producing stamens, penta-carpellary ovary (IBPGR Report 1986) and produce small to medium sized sweet fruits.

The fruits develop from the base of flowers and grow attached to the stem. Fruits are spherical, cylindrical or pear-shaped in form and vary in size and shape considerably depending on variety. Ripe fruits have thin, smooth, orange-yellow to deep orange skin. The flesh is 3-5cms thick. Within the hollow centre, in a network of fibres are embedded numerous black, wrinkled seeds each covered in a gelatinous membrane (IBPGR Report 1986).

Sex inheritance in papaya is determined by a single gene locus with three alleles: **M** is dominant for maleness; $\mathbf{M^H}$ for hermaphroditism and **m** is recessive for femaleness (Samson 1980). The diploid zygote may carry any one of the following six possible allelic combinations: **MM**, $\mathbf{M^HM^H}$, $\mathbf{MM^H}$, **Mm**, $\mathbf{M^Hm}$ and **mm.** First three homozygous dominant combinations are lethal. Progeny with these alleles do not survive. Three heterozygous combinations: **Mm** - produces male (staminate) plants, $\mathbf{M^Hm}$ - gives rise to

hermaphrodite (bisexual) plants and homozygous recessive combination **mm** results in female (pistillate) plants (Samson 1980).

Due to greater demand of 'Solo' variety by papaya farmers, there is the need to develop an efficient strategy for large-scale production of hermaphrodite seeds. Self-pollination of hermaphrodite flowers ($\mathbf{M^Hm}$ x $\mathbf{M^Hm}$) is expected to produce the following combinations: 25% $\mathbf{M^HM^H}$ (lethal therefore aborted), 50% $\mathbf{M^Hm}$ (hermaphrodites) and 25% **mm** (females). It is noteworthy that any contamination from staminate pollen would alter this ratio (IBPGR Report 1986). The actual proportions of various papaya types resulting from self-pollination of hermaphrodites, has not been determined for Fiji, where 'Solo' is the only commercial papaya variety grown. The present study was therefore undertaken to determine the proportions of various types of papaya from self-pollinated hermaphrodites flowers.

3 Materials and Methods

The experiment was carried out in Rasakulu, Sigatoka Valley, Sigatoka. Hermaphrodite flower buds were covered with brown paper bags. The bags were tied securely at stalk and remained attached until fruits ripened. Six healthy and fully developed fruits were selected randomly from a large number of fruits produced through selfing. Seeds were extracted, washed thoroughly and dried under shade. Three thousand seeds were selected randomly and sowed (1 seed per pot) in 16 x 6cm plastic pots containing alluvial soil extracted from the Sigatoka River bank. All plastic pots were filled up to the top leaving about 5cm space from the top of the pots. Out of the 3000 seeds planted in pots 2633 seeds germinated and established. The pots with germinating seedlings were watered daily. When the seedlings were three weeks old, 1g of urea was added every week to each pot. Seedlings were transplanted when they were 8 weeks old.

An area of 2.5 acres (0.626 ha) was double-ploughed and ridged two weeks before transplanting. On January 8th, 2001 these 2633 seedlings were transplanted in field. Each plastic pot was slit on the sides and placed in 20 cm wide and 30cm deep pits. Fertiliser - NPK (Nitrogen 40g, Phosphorous 30g and Potassium 30g per hole) was sprinkled in the soil prior to transplanting the seedlings. Seedlings were planted at a distance of 2 m x 2 m to ensure proper growth of plants and easy management. The plants did not require watering due to the wet season. Weeding was done around the plants every fortnight and 1g of urea added at the same time. Plants started flowering four months after planting in the field. Flowers on each of the 2633 plants were observed, identified and recorded for their sex.

4 Results

Hermaphrodite, pistillate and staminate plants were only distinguishable after four months of planting (six months after seed sowing), at the onset of flowering. Flowers of 'Solo' hermaphrodite, pistillate and staminate plants differ in size and shape. The numbers of female, hermaphrodite and male plants observed are shown in Table 1. This showed hermaphrodite : pistillate (ignoring 12 male) plants ratio 2.34:1.

Table 1 Number of hermaphrodite, female and male plants observed in field

Plant Type	Number	Percentage	Ratio Hermaphrodite: Pistillate
Hermaphrodite	1838	69.8	2.34 : 1
Pistillate	783	29.7	2.34 : 1
Staminate	12	0.5	chance occurrence
Total	**2633**	**100**	**2.34 : 1**

Table 2 below shows chi-squared (χ^2) test conducted to calculate χ^2 value.

Table 2 Chi-squared (χ^2) Test

Number	Hermaphrodite	Pistillate	Total
Observed (O)	1838	783	2621
Expected (E)	1747.3	873.7	2621
O – E	90.7	–90.7	___
$(O - E)^2$	8226.49	8226.4	___
$(O - E)^2 / E$	4.48	10.5	14.9

The offspring of 2633 self-pollinated hermaphrodite seedlings transplanted and survived in the field and evaluated for their sex ratio, produced 1838 hermaphrodite (bisexual), 783 female (pistillate) and 12 male (staminate) plants. The hermaphrodite seeds segregated in the ratio of 2.34 hermaphrodite (bisexual) : 1 female (pistillate) ignoring 12 male plants. It shows that there were more than two-third hermaphrodite plants and less than one-third pistillate plants. Staminate (male) plants were very few (only 12 out of 2633).

5 Discussion

The present study showed that selfing of hermaphrodite plants resulted in ratio of 2.34 hermaphrodite plants to 1 pistillate plants. Null (Ho) hypothesis of this ratio was tested using chi-squared (χ^2) test against a ratio of 2 hermaphrodite : 1 pistillate reported previously (IBPGR Report 1986; Aquilizan, 1987; Samson 1980). Calculated chi-squared (χ^2 0.05) value was too high (14.89) against tabulated value at 1 degree of freedom (3.84). Therefore Null hypothesis could not be accepted meaning thereby that the observed ratio of hermaphrodite plants to pistillate plants is not in agreement with reported value of 2:1 by previous researchers (IBPGR Report 1986). This deviation in observed ratio in the present study could be explained on the basis of seed selection. Although we took every care to select seeds from seed lot randomly, but most probably inadvertently we might have ended up in selecting more healthy seeds. These healthy seeds should have given rise to more hermaphrodite plants than to pistillate plants. This assumption needs to be tested to confirm this possibility. Another possibility is the presence of modifier factors or polygenes. The presence of the polygenes with small but additive effect would have changed the ratio in favour of more hermaphrodites than pistillate plants. According to the law of segregation, self-fertilised hermaphrodite plants will produce only hermaphrodite and pistillate progeny because the allelic combination for hermaphrodite is heterozygous ($\mathbf{M^Hm}$) and upon selfing will segregate into 25% lethal ($\mathbf{M^HM^H}$), 50% hermaphrodite ($\mathbf{M^Hm}$) and 25% pistillate (**mm**) combinations in F_2 generation (IBPGR Report 1986). No male offspring (**Mm** combinations) were supposed to be in the progeny of selfed hermaphrodite plants. Male (staminate) plants can only occur in the offspring when one of the parents is male. The presence of 12 male plants may have been a chance occurrence. Hermaphrodite plants are also known to change sex during long cool nights and short days (Samson 1980). Another factor could be pollen contamination from a male plant, however, this was highly unlikely because there was no male plant in the immediate vicinity of the field during bagging. The nearest papaya planting was at least 200 meter away from the experimental plot and papaya is not wind pollinated. Normally vectors (insects) visit and pollinate papaya flowers.

6 Acknowledgements

The authors wish to thank Research Station Manager and staff of Sigatoka Research Station, Sigatoka and landowner of Sigatoka Valley for their kind help in seed production and conducting field trial. Authors also wish to thank the University Research Committee for providing financial help to conduct this research work.

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