International Journal of Biology; Vol. 4, No. 3; 2012
ISSN 1916-9671 E-ISSN 1916-968X
Published by Canadian Center of Science and Education

Studies on Pollen Viability Heterosis in Parents and F_1 Hybrids of Genus *Solanum* L. (Solanaceae)

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Received: May 7, 2012 Accepted: May 29, 2012 Online Published: June 26, 2012

doi:10.5539/ijb.v4n3p117 URL: http://dx.doi.org/10.5539/ijb.v4n3p117

This research was partly financed by International Islamic University Malaysia (IIUM). Grant (TYPE A) EDW A10-155-0702

Abstract

Five collections of Solanum namely: S. melongena (purple variety, SM-1), S. melongena (white variety, SM-2), S. gillo (variety, SG-3), S. gillo (variety, SG-4), and S. gillo (variety, SG-5), were crossed in all possible combinations and the hybrids evaluated for heterosis. The results revealed percentage pollen viability was 82.3, 83.3, 67.0, 42.5 and 64.5% respectively for SM-1, SM-2, SG-3, SG-4, and SG-5. The hybrids resulting from inter varietal crosses, SM-1 x SM-2; SM-2 x SM-1 (a reciprocal cross), and SG-4 x SG-5 had 65.90, 66.3, and 76.0% respectively; while the inter specific crosses SM-1 x SG-5, SG-4 x SM-2, and SM-2 x SG-5 had 21.2, 55.5, and 63.7% respectively. The highest percentage pollen viability was recorded in SM-1 and SM-2, the two collections of *Solanum melongena*; pollen viability was also high in their reciprocal hybrids. The highest value among the hybrids, was recorded for SG-4 x SG-5 (76%), a cross between two collections of *Solanum gillo*. The lowest values (21.2% and 55.5%) were obtained for the interspecific crosses SM-1 X SG-5 and SG-4 X SM-2 respectively. One way analysis of variance at $p<.05$ revealed significant differences across studied pollens of parents and F_1 *Solanum* accessions at $p<0.000$. The inter varietal cross SG-4 x SG-5 and the inter specific cross SG-4 x SM-2 with 22.5 and 2.0% heterosis effects respectively, possessed qualitative pollen traits and could enhance effective pollination, high seed and fruit sets in subsequent generations.

Keywords: solanum, pollen viability, heterosis, interspecific crosses

1. Introduction

1.1 Background

Garden egg also known as eggplant, aubergine or brijal belongs to the family solanaceae and genus *Solanum*. The genus is tremendously large consisting of both tuberosum and non tuberosum types of species. Hunziker (1979) reported there are about 1500 species of the genus out of which1000 occurred in South America.

1.2 Description

It is an annual plant growing between 40-150cm in height and bears large coarsely lobed, leaves 10-20cm long and 5-10cm broad. The flowers are white to purple with a five lobed corolla and yellow stamens. The fruit is a fleshy berry, less than 3cm in diameter on wild plant but much larger in cultivated forms, and contains numerous small soft seeds. Wild-type variety can grow larger to about 225cm in height with large leaves over 30cm long and 15cm broad (Diggle, 1991).

In Nigeria, a large number of *Solanum* species, possibly indigenous and easily recognized by different sizes and shapes of their fruits are widely grown. Nsowah (1969) compared the yields of local and exotic egg plants and reported that one of the factors which limit productivity of the local egg plant is the poor fruit yield.

1.3 Nutrient Content

Eggplant is a good source of minerals and vitamins, and thus very useful for human nutrition. Özkaya and Dündar (2009a, b) earlier reported measures to curtail nutritional losses in fruits and vegetables. Nutrition from fruits and vegetables are significant preventive means against several diseases.

1.4 Pollen and Pollination

Asma, (2008), reported pollination plays an inevitable role in fertilization and consequently seed and fruit sets in spermatophytes, deficiency in pollen production could have direct effects on seed viability and thus fruit yield.

Cross pollination between species of this genus could facilitate better heterosis thereby developing improved line(s) with hybrid vigor for higher fruit yield and improved taste.

Apparently flower pollens can vary in size from typically 3 micrometer (as in Alpine forget-me-not, *Myosotis alpestris*) to 200 microns (cucumber flower, *Curcurbita pepo*). It is interesting to observe under light microscope that pollen grains in different plant species possessed varying dynamic shapes, distinctive body morphologies and orientations, and germinal pores (Dave, 1999).

1.5 Viability and Germinability in Pollens

Viability in pollen grain could be affected by varying factors which include temperature (Giordano, Aragão and Boiteux, 2003), humidity/ low temperature (Chira, 1963), light (Goss, 1971), and plant nutrition (Howlett, 1936).

Dafni (1992), Kearns and Inouye (1993) reported pollen grain viability could be evaluated using different efficient techniques. Pollen is viable if it has the capability to germinate on stigma of a gynaecium of related plants. The pollen upon germination produces a tube which penetrates the style and enters an ovule through the micropyle to releases two male nuclei (sperm), at this stage the micro gametophyte is matured. The two sperm effect double fertilization with the egg-nucleus on one hand and with the two polar nuclei of the central cells on the other hand. However, Sari-Gorla, Mulcahy, Villa and Rigola, (1995), earlier stated that the ability of pollen tube to emerge from the pollen coat may be regarded as an indicator of the pollen quality.

The need to boost agricultural output through production of high pollen profile, improved seed sets and high fruits (Smith-Huerta & Vasek, 1984) becomes inevitable especially to cater for the rising Nigerian population.

1.6 Aim of the Research

This research investigated pollen grain viability in selected species and the F_1 hybrids of genus *Solanum*.

2. Materials and Method

The experimental materials consisted of five collections of *Solanum* fruits. Their collection codes, identification, locality and description are shown in Table 1.

Table 1. Showed the collection codes, identification, locality and description of the materials used in this study

COLLECTION CODE	IDENTIFICATION	LOCALITY	DESCRIPTION
SM-1	*Solanum melongena*	Kwara hotel Area, Ilorin	Large, elongated and fat fruit purple in colour
SM -2	*Solanum melongene*	Saw-mill Garage Area Ilorin	Large, elongated fruit, white in colour
SG-3	*Solanum gillo*	Emir's Road, Ilorin	Small globose fruits, white in colour with green stripes
SG-4	*Solanum gillo*	Esie, Via Oro	Small round fruits, white in colour with green stripes.
SG-5	*Solanum gillo*	Central Market, Jos	Medium sized, round and lobed fruit, green in colour

2.1 Seed Extraction from Collected Solanum Fruits of Different Accessions

Seeds were carefully removed from each fruit type and mixed with fine ash, to absorb the liquid mixture of the fruit and air-dried. The dried seeds were then removed from the ash by sieving the ash. These seeds were then raised in the nursery and eventually seedlings were transplanted to the pots filled with rich loamy soil.

The experiment was carried out in an isolated area in front of plant biology department of the University of Ilorin. Five plants were raised for each variety in a row. The plants were mulched using dry grass in order to conserve moisture. Watering was done twice a day throughout the period of the experiment.

2.2 Harvesting of Pollens from Matured Flowers

The plants started flowering by the 12th week of sowing and the flowers were harvested per plant to remove the anthers. The anthers are yellowish in colour, coarse and bilobed with short filament by which they attach to the flower receptacle. The anthers open by pores at the tips of the free end. They were dabbed indoor on clean microscope slides, and the appearance of fine powder on the slides confirmed successful pollen transfer.

2.3 Staining Technique

A drop or two of Aniline Blue in Lactophenol (Maneval, 1936), was added to pollen grains on slide, and then a clean cover clip was gently lowered on stained pollen grains in such a way as to avoid splashing and air trapping. Prepared slides were observed under the microscope at x40.

Pollen grains were recorded as being viable, if they were round, deeply and uniformly stained, according to Olorode and Baquar (1976).

2.4 Hand Pollination

Equally, by hand pollination parent plants were crossed in all possible combinations by transferring pollens from anther of a plant to the stigma of another. In this way crosses were made within and between varying populations of the two species. Pollinated stigmas were encapsulated using locally made small plastics in order to prevent interference of foreign pollens. Light tags hung with threads were correctly labeled according to crosses and then hung unto pollen receptive plants. There were reciprocal crosses as well.

2.5 Pollens from Hybrids

Developed fruits were harvested separately and labeled correctly, the seeds were extracted using the method above and then sown to raise the hybrids. When the hybrids developed and produced flowers the flowers were collected and the pollens were also studied.

Data were collected in all cases, percentage pollen viability was determined for the parents and hybrid plants. The hybrids were evaluated for heterosis. One way analysis of variance was conducted using the version 16 SPSS.

3. Results

3.1 Viability Study in Pollens of Parents and F_1

The pollen data for all the collections and some F_1 hybrids are summarized in Table 2.

The percentage pollen viability was 82.3% for SM-1; 83.3% for SM-2; SG-3 had 67.0%; SG-4 had 42.5%; while SG-5 had 64.5%. The hybrid SM-1 x SM-2 had 65.90%; SM-2 x SM-1 (a reciprocal cross) had 66.3%; the hybrid SG-4 x SG-5 had 76.0%; it was 21.2% for the inter-specific cross SM-1 x SG-5; the hybrid SG-4 x SM-2 had 55.5%; while SM-2 x SG-5 had 63.7%.

The highest percentage pollen viability was recorded in SM-1 and SM-2, the two collections of *Solanum melongena*. Pollen viability was also high in their reciprocal hybrids. The highest value among the hybrids, was recorded for SG-4 x SG-5 (76%), a cross between two collections of *Solanum gillo*. The lowest values (21.2% and 55.5%) were obtained for the inter-specific crosses SM-1 X SG-5 and SG-4 X SM- 2 respectively (the crosses between *melongena* and *gillo* species).

Table 2. Percentage pollen viability for parents and hybrids of eggplants

S/N	PLANT TYPE	NUMBER OF VIABLE POLLEN	NUMBER OF NON- VIABLE POLLEN	NUMBER ON WHICH ESTIMATE WAS BASED	PERCENTAGE (%) VIABILITY
1	SM -1	1,962	422	2,384	82.3
2	SM -2	5,252	1,052	6,304	83.3
3	SG-3	898	443	1,341	67.0
4	SG-4	804	1,087	1,891	42.5
5	SG-5	2,489	1,371	3,860	64.5
6	SM -1 x SM -2	1,438	743	2,181	65.9
7	SM -2 x SM -1	926	470	1,396	66.3
8	SG-4 x SG-5	1,947	614	2,561	76.0
9	SM 1 x SG-5	154	572	726	21.2
10	SG-4 x SM -2	901	720	1,621	55.5
11	SM -2 x SG-5	1,154	654	1,813	63.7

3.2 Heterosis

The percentage heterosis obtained from both inter-varietal and inter-specific crosses were placed in Table 3, while Figure 1 showed graphical expression of the % pollen viability. The heterotic effects across the parents and hybrid varieties were shown in Figure 2.

Table 3. Estimate of % heterosis in pollen grain viability observed in the F_1 (egg plant)

S/N	PLANT TYPE	NUMBER OF VIABLE POLLEN	NUMBER OF NON- VIABLE POLLEN	NUMBER ON WHICH ESTIMATE WAS BASED	PERCENTAGE (%) VIABILITY
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11	SM -2 x SG-5	1,154	654	1,813	63.7

Key-H%: percentage heterosis; MP: Mid parent mean.

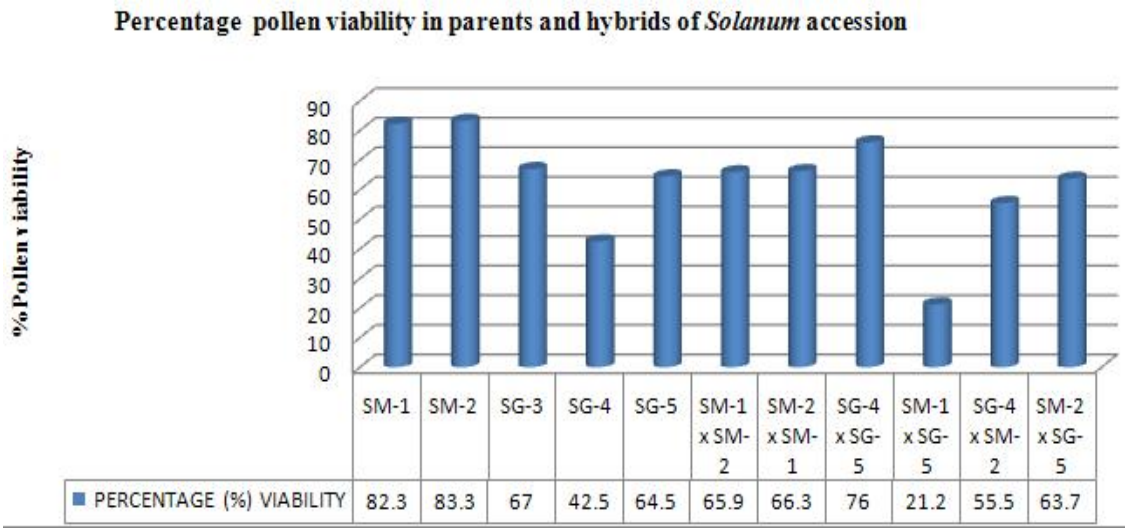

Figure 1. Showed percentage pollen viability in parents and hybrids of *Solanum* accessions

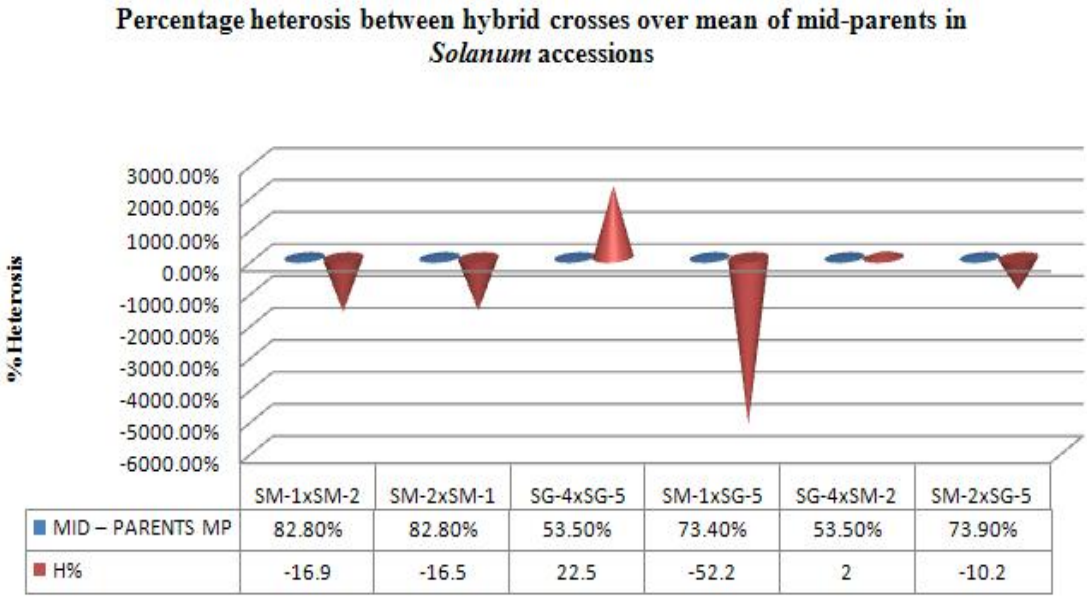

Figure 2. Showed percentage heterosis beween hybrid crosses over mean value of mid-parents in *Solanum* accessions

Analysis of variance at $p<.05$ revealed there was significant difference across studied pollens in parents and F_1 *Solanum* accessions at $p<0.000$.

4. Discussion

4.1 Heterosis

The data collected on pollen viability is shown in Table 2, while Table 3 showed the estimates of % heterosis in pollen grain observed in the F_1 Egg plants. The highest percentage of pollen viability was recorded SM-2 with 83.3%, this is followed SM-1 with 82.3. The SM-2 and SM-1 are two varieties to *Solanum melongena*. The SG-3, SG-4 and SG-5 are all varieties of *Solanum gillo* with 67%, 42% and 64.5% respectively. This result manifested in the fruit set per plant. The varieties of *Solanum melongena* and *Solanum gillo* produced fruits, with fertile and high number of seeds, throughout their life span.

4.1.1. Inter-varietal Crosses

Inter-varietal hybrids (SM-1 X SM-2 and SM-2 X SM-1) had lesser % viability when compared to the mean of the putative parents. This is simply due to inbreeding depression because the crossing involves individuals of the same population. When there is reduction in fitness in a given population as a result of breeding of related individuals, then inbreeding depression might have set in. Breeding between closely related individual results in manifestation of more recessive deleterious traits. The drop in the percentage pollen viability of the inter-varietal crosses may not be unconnected with the above reason. The more closely related the breeding pair is, the more homozygous deleterious genes the offspring may possess, resulting in very unfit individuals (Michael, 2012)

The cross SG-4 x SG-5 with higher percentage pollen viability (76.0%) over the mean of the two parents (53.5%), may possess high genetic variability within the population. Generally speaking, populations with more genetic variation do not suffer from inbreeding depression (Michael, 2012).

As a general rule, hybrid vigor (another way of saying reduction of inbreeding depression) is strongest in first generation hybrids and get weaker over time. This is what might have occurred in the inter-varietal hybrid, where the cross SG-4 x SG-5 had a higher percentage pollen viability than the parental mean. The cross SG-4 x SG-5 may be more or less hybrid of the first generation while the other crosses SM-1xSM-2 and SM-2xSM-1 may be hybrids of subsequent generations other than the first (Michael, 2012).

4.1.2 Inter-specific Crosses

The two inter-specific crosses, SM-1 x SG-5 and SM-2 x SG-5, had 21.2 and 63.7%, respectively, of percentage pollen viability. These results showed that each of the crosses had lesser % pollen viability in contrast to means of their respective parents. The two crosses involved breeding pairs from different populations. The crosses at hand are out breeding in nature and perhaps out breeding depression had set in (Michael, 2012).

This kind of depression observed in the two inter-specific crosses is said to occur when offspring from crosses between individuals from different populations have lower fitness than progeny from crosses between individual from the same population. The reason for the observed depression may be due to the breakdown of biochemical or physiological compatibilities between genes in the different populations. In the hybrid SM-1 x SG-5 for instance, the SM-1 individual from the *melongena* population will tend to have genes selected for the quality of combining well with gene combinations common in the same population; while genes found in the *melongena* population will not have selected for the quality of crossing well with genes common in the *gillo* population. Thus out breeding can undermine vitality by reducing positive epitasis and / or increasing negative epitasis.The obtained results are in line with previous findings by Omidiji (1981); and Scaff, Jelenkovic, Boyer & Pollack, (1982) in crosses involving *Solanum* species.

The out crossing SG-4 x SM-2 is the only hybrid where the percentage pollen viability (55.5%) was higher than the parents mean. Though this cross is inter-specific, its ability to possess pollens with higher percentage viability than the mean of the two parents may be due to breakdown in the co-adapted gene complexes that usually set in during outbreeding. Since as a general rule, outbreeding can be relatively weak in the first generation, but will increase in power through further generations as co-adapted gene complexes are broken apart without forging of a new one to take their place (Michael, 2012).

It is possible that either of, or the two parents (SG-4 and SM-2) used in this work might be hybrid(s) of previous crossings, such that the SG-4 x SM-2 cross is a further generation of the previous ones, hence the better % pollen viability.

5. Conclusion

The inter varietal cross SG-4 x SG-5 and the inter specific cross SG-4 x SM-2 with 22.5 and 2.0% heterosis respectively, have qualitative pollen traits that would enhance effective pollination, high seed and fruit set in subsequent generations.

Acknowledgement

We are thankful to the research management center of the International Islamic University Malaysia (IIUM) for partly sponsoring this research through the endowment fund, 'EDW A10-155-0702' JAZAKUMULLAHU KHAIRAH.

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