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Floral biology and phenology of sweet potato (*Ipomoea* batatas [L.] Lam.) in Cuba: Bases for genetic improvement

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Despite the fact that some aspects related to the flowering of the sweet potato have been studied previously, to date, in Cuba and tropical regions, only general descriptions have been documented without detailed information on the biology and floral phenology of the sweet potato, aspects of interest for genetic improvement. To carry out the research, four sweet potato genotypes obtained and released by the genetic improvement program of the Research Institute of Tropical Roots and Tuber Crops (INIVIT) were selected, one of them ("CEMSA 74-228") is of international reference. The average flower emission frequency per inflorescence was 1.5 days. The number of flowers per inflorescence fluctuated between 7.35 and 26.14. The time from pollination to seed maturation varied on average from 22.8 to 32.3 days. For the four genotypes, different moments of anthesis were found, ranging from 0444 h (\pm 8.3 min) to 0619 h (\pm 10.7 min). The pollen presented viability percentages above 90% from 0600 to 1300 h. The results allowed a better knowledge of the floral biology of the sweet potato and the floral phenological phases and the approximate time of each one was determined, which will be useful for plant breeders

Key words: Biology, flowering, phenology, *Ipomoea batatas*, pollination.

INTRODUCTION

Convolvulaceae is a family that includes approximately 2000 species and 58 genera distributed throughout the world, mainly in the tropics and subtropics (Staples and Yang, 1998; Staples, 2011; Wood et al., 2020). More than a third of the species are included in two main genera, *Ipomoea* and *Convolvulus* (Carlquist, 1988). The

genus *Ipomoea* Linnaeus it represents 600 to 700 species distributed in the world, although more than half of them are concentrated in the Americas (Austin and Huamán, 1996; Mabberley, 1997). Within the sweet potato series there are 13 wild species that are considered to be closely related to the sweet potato

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Author(s) agree that this article remain permanently open access under the terms of the <u>Creative Commons Attribution</u> <u>License 4.0 International License</u> (Austin, 1978, 1979).

Linnaeus (1737) described the sweet potato and assigned it its binomial nomenclature, designating it as Convolvulus batatas L. In 1791, Lamark considering the morphology of the stigma and pollen grains (genus Ipomoea: capitate stigmas and generally spiny pollen grains) changed the genus from Convolvulus to Ipomoea and definitively designated this species as Ipomoea batatas (L.) Lam. (Montaldo, 1991; Huamán, 1999). Sweet potato is an autopolyploid (2n=6x=90, x=15), a highly heterozygous clone hybrid (Grüneberg et al., 2015), with easy generation of true seeds (in Cuba) through directed crossing and open pollination (a successful pollination result in 1-3 true seeds), in the latter the main pollinating agent are bees (Morales et al., 2017). The plant has a sporophytic self-incompatibility system (Martin and Cabanillas, 1966; Veasey et al., 2007; Silva et al., 2012; Grüneberg et al., 2015; Alves et al., 2017: Grüneberg et al., 2022).

Flower formation of sweet potato, is in general, related to latitude; it flowers frequently in the tropics while little or no flowering occurs in temperate regions. Even in the tropics, the intensity of flowering varies widely with seasons from poor to abundant, and some varieties produce little or no flowers at all (Schreven, 1954; Rheenen, 1965). It is generally recognized that a short photoperiod and a slight humidity, promote sweet potato to flower. Most of the sweet potato genotypes flower naturally in the short days in the tropics in Cuba from November to April (Morales et al., 2017) and it is not necessary to use grafts on *Ipomoea* spp. to induce flowering (Edmond and Ammerman, 1971).

In Cuba, since 1967, a sweet potato Genetic Improvement Program (GIP) has been developed, which applies the principles of genetics to produce new cultivars, with superior characteristics, such as higher yield, resistance to biotic and abiotic factors, higher nutritional values, etc. Annually in the hybridization campaign, the blocks of crosses with the selected parents of different genetic constitution are designed to meet the objectives set. Hybridizations are carried out at the Research Institute of Tropical Roots and Tuber Crops (INIVIT) (Morales et al., 2017). Currently, 40 000 hybrid seeds are produced annually and in total more than one million seeds have been obtained for uses in genetic improvement.

Aspects related to sweet potato flowering have been previously studied due to the importance of this species for the world (Chatterjee and Nagbiswas, 1952; Edmond and Ammerman, 1971; Jones, 1980; Hernandez, 1995; Huaman, 1999; Montaldo, 1991; Lebot, 2010; Martí et al., 2014; Reddy et al., 2018; Brito et al., 2021; Ozturk, 2021; Behera et al., 2022; Shankar and Kaushik, 2022). However, to date, in Cuba and tropical regions, only general descriptions have been documented without detailed information on the biology and floral phenology of the sweet potato, aspects of interest for genetic improvement.

The production of hybrid seed in sweet potatoes by directed crosses is expensive, very laborious and requires intense technical supervision. Given the importance of hybridization as a genetic improvement strategy and the lack of information regarding the floral biology of *I. batatas* in tropical regions, this research aimed to reveal the main aspects related to sweet potato floral biology and phenology. To carry out the research, four sweet potato genotypes obtained and released by the INIVIT GIP were selected, one of them, "CEMSA 74-228" released in 1974, is an international reference (Tique et al., 2009; Nawal and Gayoum, 2012; Amoatey et al., 2016; Abidin et al., 2017; Swanckaert et al., 2020; Grüneberg et al., 2022; Nakitto et al., 2022). The result of the study will allow a better knowledge of its floral biology and establish the approximate time of its phenological phases based on the characteristics of the parents, this will help fill the knowledge gap and will be useful for plant breeders.

MATERIALS AND METHODS

Study area

The experiment was carried out at the Research Institute of Tropical Roots and Tuber Crops (INIVIT), Santo Domingo municipality, Villa Clara province, Cuba, located at 22°35'00"N and 80°14'18"W, at 50 masl. The experiment was carried out under field conditions during the months of October 2022 to April 2023 in a soft carbonated brown soil (Hernández et al., 2015). Crossing blocks were planted, using for the study four sweet potato genotypes: "CEMSA 74-228", "CEMSA 78-326", "INIVIT BS-16", and "INIVIT BM-90", with 50 plants per genotype at a distance of 1×1 m, with a cutting of the same clone in each planting position. In each planting position, a 2 m high stake was placed and tied at the top. Meteorological data were recorded at the automatic meteorological station belonging to the institute (national meteorological network code: 78326, data available at: http://www.insmet.cu).

Morphometric characters

In the month of December, a morphometric characterization of 30 flowers of the four sweet potato genotypes was carried out. Flowers were collected at 0800 h and photographs were taken with a professional CANON EOS T100 EF-S-18-55 camera. The characters evaluated were the following: Corolla length (CL), Corolla width (CW), Pistil length (PiL), Tallest stamen length (TSL), Length of inner sepals (LIS), and Pedicel length (PeL). The measurements were made with the professional software for digital image analysis programmed in Java: ImageJ ver. 1.46 from National Institute of Health, following the instructions of Ferreira and Rasband (2012).

Growth of the inflorescence

In order to determine the development of the inflorescence, six weeks after planting, 30 inflorescence emissions per genotype were marked. Daily visual monitoring was carried out for 3 months and the length of the inflorescence, emission frequency of flower per inflorescence and total number of flowers emitted per inflorescence

(NFI).

Number of flowers per plant (NFP)

In the crossing blocks, the total number of flowers per plant was counted. 30 plants were selected by genotypes and the number of flowers emitted was recorded daily. The count was carried out for five and a half months (November 2022-April 2023).

Time from pollination to seed maturation (TPSM)

In the month of December, 100 flowers were pollinated per genotype and directed biparental crosses were carried out between clones of the same compatibility group: CEMSA 74-228 \times INIVIT BS-16 and CEMSA 78-326 \times INIVIT BM-90. The selected flowers covered the day before anthesis by their apical end with a small section of Smoothie Straws, the time selected for this activity was between 0800 and 1000 h. After pollination, the flowers were covered again, labeled and the capsules were harvested once the pedicel was totally necrosed according to the Jones (1980) methodology.

Moment of anthesis (Ant)

To determine the exact time of anthesis, eight plants per genotype were selected, which were filmed 10 times each, on selected days of December, January, and February. A Sony HDR-PJ790V camcorder with night vision system was used, which was placed on a tripod at a distance of approximately 1 m from the flowers. Each flower was filmed for 32 h, from 0800 h (day before anthesis) to 1600 h (days after anthesis). Flowers were considered open when the distal end of the corolla opened, even when not fully extended.

Number of pollen grains per anther

To count the number of pollen grains per anther, the flowers were collected at 0300 h (before the anther dehiscence). Twenty flowers were collected per genotype (100 total anthers per genotype), the anthers were placed separately on slides. Once the anthers dehisced, with the help of entomological pins and tweezers, the pollen grains were extracted, which were stained with 1% acetocarmine. Under a stereoscopic microscope (Zeiss 2000-C), we proceeded to count the pollen grains.

Pollen viability and stigmatic receptivity

To determine the viability of the pollen, 50 flowers were randomly collected and the pollen was extracted with the help of tweezers. Staining with 1% acetocarmine was used; the instructions of Ordoñez et al. (2017) were followed. Pollen grains that were stained are viable, while those that remain uncolored are non-viable. Counting was performed with the help of a stereo microscope (Zeiss 2000-C) to determine the percentage of viability. 10 measurements were made during the course of the day, from 0600 to 1600 h, with 1 h intervals.

Receptivity of the stigma was evaluated using the drop hydrogen peroxide test (H_2O_2) at 3% (Zeisler, 1933). To achieve this, 10 µL of H_2O_2 was applied on the stigma with a micropipette (Micropipette 1–10 µL), after which the observations were carried out with the aid of a stereo microscope (Zeiss 2000-C). Twenty Stigmas were evaluated, evaluations were performed at 72, 48, and 24 h before anthesis and from 0600 to 1600 h after anthesis, with 1-h intervals. Depending on the level of bubbles produced, a percentage value

was assigned to each observation. In all cases, stigmas of recently collected flowers were used.

Pollen viability and stigmatic receptivity were only measured on the "INIVIT BS-16" genotype.

Statistical analysis

In order to analyze the evaluated variables, the data was tabulated and organized in an Excel matrix, the means and standard error were used as parameters of the descriptive analysis. To test the normality of a dataset, the test of Kolmogorov-Smirnov was used. An Analysis of Variance (ANOVA) was performed and when there were significant statistical differences, the means were separated using Duncan's multiple range procedure (α =0.05).

For the continuous quantitative variables TNFI, NFP, TPSM and Ant, a Principal Component Analysis (PCA) was performed in order to know which ones are or not associated. Its eigenvector matrix (coefficients of the linear combinations of the original variables) was interpreted. With the correlation values of the variables, their projection on the first two axes (first and second dimension) was represented.

The relationship between the variables NFI (independent variable) and TPSM (dependent variable) was determined through a linear regression. Software was used RStudio in R programming language (Hernández and Usuga, 2023).

RESULTS

Parts of the sweet potato flower, capsule and seed

Sweet potato flowers are hermaphroditic, composed of five stamens (male organs or androecium) and a pistil (female organ or gynoecium). Pollen grains are spherical and with the surface covered with glandular hairs. The stigma is bicapitated. The style ends in a broad stigma and is covered with glandular hairs. The base of the ovary is yellow, which contains nectar. The calyx consists of five sepals, two outer and three inner. The fruits are spherical capsules with a terminal tip, glabrous or hirsute and dehiscent; once mature they turn brown. This is a general description for the four genotypes evaluated (Figure 1).

Morphometric variables

The examination of the p-values of all the morphometric variables evaluated shows that there are significant statistical differences (p-value < 0.05) between the means of the genotypes at a significance level of 0.05. The length of the corolla varied between 4.60 and 2.83 cm. The highest value of corolla width (5.12 cm) was found in the "INIVIT BM-90" genotype with statistical differences from the rest of the genotypes. Both the length of the pistil and the length of the tallest stamen were variable by genotype, thus evidencing different degrees of heterostyly in the flowers. The length of the inner sepals varied between 0.95 and 1.15; there were significant differences between the "CEMSA 74-228" genotype (with the highest value) and the rest of the



Figure 1. Parts of the sweet potato flower, capsule and seed; cultivar: CEMSA 78-326.

genotypes. The highest values of pedicel length were found in the "CEMSA 74-228" and "INIVIT BS-16" genotypes with 1.20 and 0.74 cm, respectively (Figure 2).

Inflorescence growth, flower and emission frequency per inflorescence, number of flowers per inflorescence, and number of flowers per plant

The growth of the inflorescence peduncle was measured from the activation of the axillary bud until 26 days later. The maximum growth reached between the genotypes varied between 5.3 and 10.2 cm, this growth occurred until 20 days later, from which time the growth in length stopped. This coincides with the emission of the first flower of the inflorescence, which occurred between 18 and 20 days. The frequency of emission of flowers per inflorescence average per genotype was 1.5 days and that an inflorescence remains emitting flowers was different by genotype, for "CEMSA 74-228", it was 35.8 days, in "CEMSA 78-326" it lasted 15.2 days, in the "INIVIT BM-90" 17.3 days and in the "INIVIT BS-16" 12.5 days. Two genotypes ("CEMSA 74-228" and "INIVIT BM-90") emitted two flowers per inflorescence on the same

day; the percentage probability for this to occur was 33.3% for "CEMSA 74-228" and 20.0% for "INIVIT BM-90" (Figure 3).

The examination of the p-values in the number of flowers per inflorescence shows that there are significant statistical differences (p-value < 0.05) between the means of the genotypes at a significance level of 0.05. The highest number of flowers per inflorescence (26.14) was found in the "CEMSA 74-228" genotype with statistical differences with the rest of the genotypes and the lowest value (7.35) was found in the "INIVIT BS-16" genotype. The total number of flowers per plant varied by genotype, with significant statistical differences between them (p value < 0.05); varying between 758.8 (with a maximum value, "CEMSA 74-228") and 434.0 (minimum value, "CEMSA 78-326") (Figure 3).

Time from pollination to seed maturation

The examination of the p-values for the time from pollination to seed maturation shows that there are significant statistical differences (p-value < 0.05) between the means of the genotypes at a significance level of



Figure 2. Morphometric variables.

0.05. This time varied as a general average between 22.8 and 32.3 days, with the shortest time being the "CEMSA 78-326" genotype and the longest being "CEMSA 74-228". It was found that once the pedicel turns yellow, it takes two days to become completely necrotic, at which time the capsules are ready to be collected (Figures 4 and 5).

Moment of anthesis and number of pollen grains per anther

For the four genotypes, different moments of anthesis were found. The first genotype that anthesis occurred

was in "CEMSA 78-326" at 0444 h (\pm 8.3 min), taking into account that at that time of the year dawn occurs at 0630 h, in this cultivar anthesis occurred 1:46 h before dawn, then in the "INIVIT BM-90" at 0538 h (\pm 7.8 min), in the "CEMSA 74-228" at 0602 h (\pm 11.2 min) and in the "INIVIT BS-16" at 0619 h (\pm 10.7 min). The difference in anthesis time between the four genotypes was 1:35 h. The results indicate a greater occurrence of anthesis in dark conditions in the early hours of the morning. The total decontortion of the flower occurred between 1:53 and 2:38 h (Figures 6 and 7).

The examination of the p-values for the number of pollen grains per anther shows that there are significant statistical differences (p-value < 0.05) between the means



Figure 3. Inflorescence growth, flower and mission frequency per inflorescence. Number of flowers per inflorescence and number of flowers per plant. Source: INIVIT (2023).



Figure 4. Time from pollination to seed maturation; a: Different stages of the flower in the inflorescence. Source: INIVIT (2023).

of the genotypes at a significance level of 0.05. The "CEMSA 74-228" genotype presented 461.1 pollen grains per anther, the "INIVIT BS-16" 440.5 and the "INIVIT BM-90" 444.7, there being no significant statistical differences between these genotypes, although there were between

these three with the genotype "CEMSA 78-326", which presented 359.5. Considering the average number of pollen grains per anther among the genotypes (426.4); it can be presumed that a sweet potato flower is capable of producing 2132.2 grains of pollen and a sweet potato



Figure 5. Sequence from pollination time to seed maturation, "INIVIT BM-90" genotype. Number mean days.





plant around 1360375.5 (Figure 6).

Pollen grain viability and stigmatic receptivity

The pollen presented viability percentages above 90%

from 0600 to 1300 h, after that moment until 1500 h it also presented a high viability, ranging from 80 to 92% (Figure 8).

The stigmatic receptivity was below 25% in the measurements made at 72, 48 and 24 h before anthesis.



Figure 7. Sequence from 0800 h (before anthesis) to 1600 h (after anthesis), "CEMSA 78-326" genotype.

Between 0600 and 0800 h, the receptivity ranged from 79 to 93%, the stigma reached 100% receptivity between 0900 and 1100 h. From 1200 h, the receptivity began to decrease until it reached 64% at 1500 h. After 1200 h, the existence of tissue damage was evidenced (Figure 8).

It was observed that the variance associated with each dimension is different and decreases in order. The first dimension explained 73.5% of the total variance and the

second dimension 12.5%. The first two dimensions together explained 86% of the variance. The projection of the correlation of the genotypes and the four variables of interest for genetic improvement (TNFI, NFP, TPSM and Ant) in the first two dimensions (Figure 9) was represented. Proximities between variables are interpreted as similar behavior of these with respect to the genotypes (close values in them); it also means that they are highly correlated with each other. The



Figure 8. Pollen grain viability and stigmatic receptivity, "INIVITBS-16" genotype. ba: before anthesis.

representation of the genotypes is translated into four well-defined clouds of point-variables (groups of variables). The variable most related to dimension one in a positive sense was TNFI and in a negative sense Ant. The directions of the variables and their angular separation showed that they all influence in the same direction (Figure 9).

The result of the linear regression between NFI (independent variable) and TPSM (dependent variable) indicated that for each additional flower in the inflorescence, TPSM can be expected to increase by an average of 1.54 days. In addition, the value of R2 = 0.47,



Figure 9. Principal component analysis (PCA) for genotypes and four variables: Total number of flowers per inflorescence (NFI), Number of flowers per plant (NFP), Time from pollination to seed maturation (TPSM), Anthesis (Ant); Linear regression for the TPSM (dependent variable) and NFI (independent variable) variables.

indicated that only 47% of the variability of the TPSM variable is explained by NFI (Figure 9).

DISCUSSION

The morphology of sweet potato flowers has been previously described by different authors; our results agree with Reddy et al. (2018), who reported that flowers, pedicel and peduncle, differ in size. The corolla 2-4 cm long (Montaldo, 1991) or 2.5-7.0 cm long (Austin, 1978), by 2-3.5 cm wide. Martí et al. (2014) indicate that sweet potato flowers are 2.8 to 6.3 cm in height and 2.6 to 5.6 cm in diameter. The edges of the mesopetal areas are purplish or violet, the interior strongly purple-reddish, although there are genotypes with totally white corollas (Austin, 1978; Montaldo, 1991).

Pistil length in our evaluations varied between 1.66 and 2.55 cm. Jones (1980) reported that stamen filaments vary in length from 5 to 21 mm between cultivars and within a plant flower, filaments can vary in length and location relative to the stigma such that any number of anthers it may be below, equal to, or above the stigma, and the pistil may be 8 to 29 mm long. Adi et al. (2013) based on the morphology of the flower of two genotypes of I. batatas, reported that the stigma length was higher than the tallest anther with relative distances 5.12 mm in one sweet potato variety and while in the other it was 4.90 mm. In our study, when comparing the length of the pistil with the tallest anther, two of the genotypes presented flowers with morpho-brevistyle (anther taller than the pistil) ("CEMSA 78-326", "CEMSA 74-228") and two morpho-longistyle (pistil higher than the anther) ("INIVIT BS-16", "INIVIT BM-90"). According to Poole (1952) and Paredes (2014), sweet potato flowers present the phenomenon of heterostyly. The phenotype for each morph is genetically determined (Ganders, 1979). Yen (1963) has classified sweet potato cultivars in terms of heterostyly into 5 groups (Heterostilic, Intermediates (3 groups) and Hemostyle).

The results of the present study indicate that the number of flowers per inflorescence can vary from 7.35 to 26.14. Chatterjee and Nagbiswas (1952) report that sweet potato flower buds are born in clusters of 7-10, rarely more. Jones (1966) reported that sweet potato flowers occur in axillary inflorescences of 1 to 22 buds that open singly or in groups. Saxena (1986) and Hernández (1995) report that sweet potato inflorescences produce 3 to 7 flowers per peduncle.

The number of flowers per plant in our research ranged from 434.0 to 758.8. Nwankwo et al. (2019) in Nigeria reported that the flower production of 14 sweet potato parents was 3 896 flower buds with a mean of 556.6 flower buds per plant. They reported that the number of flower buds may decrease due to a drastic reduction in the moisture content of the soil. Soil water stress affected the flower health of the parents, resulting in gradual senescence and death of the sweet potato plant, which invariably affected flower production. Chatterjee and Nagbiswas (1952) reported that up to three flowers on the same inflorescence opened on the same day. Our results are consistent, since two genotypes ("CEMSA 74-228" and "INIVIT BM-90") emitted two flowers per inflorescence on the same day.

Brito et al. (2021) in Brazil studied 22 sweet potato genotypes and concluded that flowering usually begins 125 days after planting and lasts for an average of 72 days. Our results do not coincide with those of the previous author, because in our tropical climatic conditions the sweet potato genotypes began to flower in the month of November approximately 45 days after planting. In Cuba, flowering is seasonal and influenced by the photoperiod (short days, long nights), regardless of the age of the plant, they begin to bloom in this month. Flowering lasted for five and a half months until April. Although, there are some accessions in the germplasm collection that flower throughout the year and others that never flower.

The results of the present study show that the time between pollination and seed maturation is highly variable by genotype. This time varied as a general average between 22.8 and 32.3 days. Jones (1980) reports that the seed is mature in about 1 month and when the capsules are completely dry and brown, the seeds are ripe.

Chatterjee and Nagbiswas (1952) observed under Delhi (India) conditions that flower buds that are half open at 2 pm will not open further until early the next morning (4 to 6 am). They suggest that the flowers buds are very sensitive to atmospheric temperature. A cold snap invariably delays the opening of flower buds. In our experiment, it was possible to observe on two occasions when the temperature at dawn and in the morning dropped below 10°C, at 1000 h the corolla had hardly expanded, remaining half-open. This is because the sweet potato stops its growth at 15°C and does not survive below 12°C (Montaldo, 1991). Jones (1980) reported that sweet potato flowers open shortly after dawn and generally wither by noon, depending on environmental conditions. On cloudy days the flowers may stay open longer than on hot sunny days. Terada et al. (2005) studied four Ipomoea species in Brazil and their results showed that Ipomoea papa flowers open at 6:00 am and close at 2:00 pm. Xiaocheng et al. (2007) studied the pollination biology in Ipomoea cairica and reported that enthesis occurred between 4:30 and 5:20 h and lasted mainly until the afternoon when the corollas withered and closed at 17:40 h. Maimoni-Rodella and Yanagizawa (2007) studied the floral biology of three wild species of Ipomoea (I. cairica, Ipomoea grandifolia and Ipomoea nil) and found that the anthesis began around 5:00 h, they also reported that at the time of floral opening, the stigma was receptive and the anthers dehiscent. Lima and Pigozzo (2013a) studied the reproductive biology of Ipomoea eriocalyx and they reported that the anthesis of the flowers occurred at 5:00 h. Lima and Pigozzo (2013b) also studied the floral biology of Merremia dissected var. Edentate (Convolvulaceae), finding that the opening of the flower buds begins at 8:00 a.m.

The observations of the present study in Cuba suggest that the dehiscence of the anthers begins simultaneously during anthesis, before the complete decontortion of the flower is completed, the anthers are dehiscent. In the present study, the wilting of flowers was influenced by temperatures, humidity and solar radiation; on the hottest and sunniest days the flowers began to wither at 1100 h. while on less hot and cloudy days they began to wither at 1100 to 1200 h. The corolla detaches from the pedicel in the early morning hours of the next day. Chatteriee and Nagbiswas (1952) found that anther dehiscence in I. batatas starts from the top down. The anthers usually dehisce 2 h after the opening of the flower. Only in 6% of the cases the anthers were dehiscent simultaneously with the opening of the flowers. They suggest that higher humidity accelerates dehiscence. The withering of the flowers occurs after 10:30 am. The petals first begin to

lose their color and fade. The fade continues to the base. At this time the shiny stigma loses its shine and begins to dry out. Later, it turns brown. The flower falls off the pedicel after 24 to 30 h of opening.

Chatterjee and Nagbiswas (1952) observed a pollen fertility is very high, ranging from 76.5 to 100%. The stigma is receptive to the sweet potato in the early hours of the morning and pollination is mainly due to bees (Montaldo, 1991; Huamán, 1999; Martí et al., 2014). The stigma receptivity test on the species *I. papa* showed that it was receptive between 0635 and 0100 h. And the highest viability of pollen grains between 0635 and 0700 h, although viability is maintained throughout the day until the flowers close (Terada et al., 2005). Xiaocheng et al. (2007) in the species I. cairica report both pollen viability and stigma receptivity remained at a high level (> 90%) during the flowering period on the first day. Adi et al. (2013) reported that pollen viability in sweet potato has a range of 70.25 to 91.52%. They observed 100% receptivity of the pistil and 80 to 100% dehiscence of the anther. Nwankwo et al. (2019) reported that the time for pollen fertility and stigma receptivity in sweet potato is high early in the morning between 0700 and 0800 h and decreases as the day progresses. In our study, the use of hydrogen peroxide identified a very strong positive response even after the stigma presented tissue damage. This method is considered inexpensive and is capable of identifying the reaction of the peroxidase enzyme present on the stigma from the air bubbles released during the reaction.

Jones (1980) reported that not much is known about the longevity and viability of sweet potato pollen because no one has successfully germinated it in culture. Adi et al. (2013) reported that they had managed to germinate them successfully. Our research results confirm what was indicated by Jones (1980), despite several attempts to germinate pollen grains and having followed the instructions and culture media proposed by Adi et al. (2013); it was not successful. This indicates that there are still gaps in the knowledge of the germination of pollen grains in sweet potato.

Considering the results of the investigation, three phenological phases are proposed for the sweet potato inflorescence, which are:

Phase 1 (Growth): Duration of 18-21 days, it begins with the activation of the floral bud in the axil of the leaf until the emission of the first flower of the inflorescence. In this phase the growth of the floral peduncle and the development of the flower buds of the inflorescence occur, at the end of this phase the growth in length of the peduncle stops.

Phase 2 (Flowering and fertilization): duration of 18-32 days (from 18-21 days to 36-53 days of age), beginning with the emission of the first flower until the emission of the last flower. In part of this stage the emission of flowers, pollination, maturation of capsules and harvest of



Figure 10. Phenological phases of sweet potato inflorescence.

seeds coincide at the same time.

Phase 3 (Maturation): Duration of 24-32 days (from 36-53 days to 60-85 days of age). It begins with the emission of the last flower until the maturation of all the capsules of the inflorescence. In this stage the maturation of most of the capsules occurs and therefore the collection of botanical seeds. In some genotypes the capsules open days after full maturation, however, in others the capsules can remain unopened for several weeks, an aspect highly influenced by solar radiation, relative humidity and temperature. It was also observed that, despite the necrosis of all the inflorescence pedicels, the floral peduncle may or may not remain green (Figure 10).

Conclusion

The results allowed a better knowledge of the floral biology of the sweet potato and the floral phenological phases and the approximate time of each one ("CEMSA 74-228", "CEMSA 78-326", "INIVIT BS-16" and "INIVIT BM-90") was determined, which will be useful for plant breeders. The maximum growth reached of the floral peduncle varied between 5.3 and 10.2 cm. The frequency of emission of flowers per inflorescence was 1.5 days. The number of flowers per inflorescence fluctuated between 7.35 and 26.14. The time from pollination to seed maturation varied on average from 22.8 to 32.3 days. For the four genotypes, different moments of anthesis were found, ranging from 0444 h (\pm 8.3 min) to 0619 h (\pm 10.7 min). The total decontortion of the flower occurred between 1:53 and 2:38 h. The pollen presented

viability percentages above 90% from 0600 to 1300 h, while the stigmatic receptivity was below 25% in the measurements made at 72, 48 and 24 h before anthesis. Between 0600 and 0800 h, the receptivity ranged from 79 to 93%.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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