EFFECT OF PISTIL AGE ON POLLEN TUBE GROWTH, FRUIT DEVELOPMENT AND SEED SET IN *CUCURBITA PEPO* L.

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ABSTRACT

The effects of pistil age on pollen tube growth, fruit development and seed set were studied in *Cucurbita pepo* L., the flower of which opens for only six hours. Stigma receptivity lasts four days, from one day before until two days after anthesis. Style receptivity lasts three days, from the day before to the day after anthesis. Ovule receptivity lasts two days: the day of anthesis and the day before. The rate of pollen tube growth varies in different parts of the pistil and in relation to pistil age. In the stigmatic and stylar region, the tubes grow faster if pollination occurs the day before anthesis; in the ovary they grow faster when pollination occurs at anthesis. In the receptacle region, where the transmitting tissue is reduced, the growth rate decreases independently of the time of pollination. The fruits are larger and heavier with more seeds when pollination occurs at anthesis. There is a positive correlation between seed number and fruit weight when pollination occurred at anthesis and the day before.

KEY WORDS: *Cucurbita pepo*, pistil age, pollen tube growth, pollen tube competition, seed set, fruit set.

INTRODUCTION

Fruit and seed set in flowering plants are affected by many biotic and abiotic factors. The main biotic factors are pistil receptivity, pollen viability and pollen load size. Pistil receptivity includes stigma, style and ovule receptivity; the period in which these three types of receptivity overlap is called the effective pollination period (EPP) (Williams 1970). For fertilisation to occur, the pollen landing on the stigma during the EPP must be viable, i.e. it must be able to germinate, penetrate the stigma and style and reach the ovule (Shivanna et al. 1991).

It has been demonstrated that the weight and size of fruit and the number of seeds they contain are directly proportional to pollen load size in various species (Mann 1943; Visser and Verhaegh 1987; Winsor et al. 1987). A low pollen load may cause fruit shape anomalies (Mann 1943). This is especially important for cultivated plants such as zucchini, the fruit of which have many seeds and the commercial value of the fruit depends largely on uniform shape and size (McGregor and Todd 1952). The presence and abundance of pollinators during the blooming of entomophilous species is therefore important.

Knowledge of the most efficient period for pollination is of use to plant breeders for optimising the number of seeds per fruit and predicting fruit size at maturity. Most studies on fruit and seed set of Cucurbitaceae of commercial interest have been performed at anthesis of the female flower,

which is usually short (6-36 hours) (Pouvreau 1984). Research on *Cucumis melo* and *Cucumis sativus* has exploited variations in pistil age to overcome pre-fertilisation barriers (van der Knaap and de Ruiter 1978) but the consequences for fruit and seed set were not analysed. Le Deunff et al. (1993) studied the effect of ovule receptivity on seed set and fruit development in *Cucumis sativus* but did not investigate the effect on pollen tube growth. Stephenson et al. (1985, 1988a, b) studied non-random fertilisation of the ovules and the effects of pollen load size on fruit maturation and sporophyte quality in *Cucurbita pepo*. Winsor and Stephenson (1995) studied pollen tube attrition in the same species.

The aim of the present study was to investigate the effects of pistil age on pollen tube growth, fruit size and seed set in *Cucurbita pepo* (Cucurbitaceae), a monoecious species in which anthesis lasts only 6 h (Nepi and Pacini 1993). The study is part of a research programme on the reproductive biology of *Cucurbita pepo*.

MATERIALS AND METHODS

Plant material

Plants of *Cucurbita pepo* cv. Tonda di Nizza (F1 hybrid) were grown in the open air in the Botanical Gardens of Siena University, Italy, in summer 1996.

Growth of ovary and corolla

Ovary and corolla growth were documented by measuring 30 flowers in each stage from ten different plants for 10 days before anthesis. Characters useful for determining pistil development, such as changes in petal colour, style elongation, stigma colour and exudate, were recorded.

Stigma receptivity

Stigma receptivity at different ages of the pistil was determined in two ways: a) by testing peroxidase activity of the stigma (Kearns and Inouye 1993); b) by detection of pollen tubes in transversal hand section of stigma stained with aniline blue (O'Brien and McCully 1981), 15 min after pollination with pollen from flowers open the same day. In the same sections, the percentage of germination of the pollen on stigmas of different age was calculated from the observation of 100 pollen grains per stigma in ten stigmas of each age.

Scanning electron microscopy

Stigmas were dissected 30 min. after pollination, fixed on stubs and coated with gold in an Edwards vaporizer. They were observed at 7.2 kV with a Philips 501 scanning electron microscope.

Pollen tube growth and receptivity of style and ovules

Female flowers were pollinated at 8 am of each day at different flower ages: from three days before to two days after anthesis. Male and female flowers were bagged the day before in order to avoid the interference of pollinators. A mixture of pollen was prepared collecting pollen from newly dehiscent anthers of different plants. Each of the three stigma lobes was saturated with pollen applied by a small paint-brush. The number of pollen grains adhering to the stigma were estimated in 20 flowers as described by Kearns and Inouye (1993). Pollen viability was tested

before hand pollination by the fluorochromatic reaction (Heslop-Harrison et al. 1984).

Pistils were removed 2, 4, 6, 24 and 28 hours after pollination to measure the position of the tubes and to determine their rate of growth in the different parts of the pistil. Pollen tube growth was evaluated by measuring the distance from the stigmatic surface reached by the three longest pollen tubes as described by Winsor and Stephenson (1995). Fifty pistils were observed for each stage of development and for each pollination time. Cross hand sections were stained with aniline blue to locate the position of the pollen tubes in the various parts of the pistil.

Pollen tube pathway

To identify the path of the pollen tubes, ten stigmas were pollinated on only one of the three lobes. At different times afterwards, cross-sections were cut, stained with aniline blue to locate pollen tubes and observed to determine whether pollen falling on a stigma lobe reaches the ovules in the same or other bundles of transmitting tissue.

Ovary growth and ripening of fruit

Two sets of 50 flowers were pollinated, as previously described, one day before anthesis and at anthesis. Ovary diameter was measured every two days until the fruits reached their maximum size. The fruits were picked and weighed; the number and weight of dry seeds per fruit were determined.

Statistical analysis

Distribution fitting and correlation analysis were carried out using Statistica for Windows (release 4.5, StatSoft Inc. 1993). Chi-square goodness of fit test for normal distribution was applied to fresh weight and number of seeds variables. Data were compared with a t-test for two samples analysis. Number of seeds and weight of fruits variables

DAYS BEFORE AND AFTER ANTHESIS	-10	-7	-3	-2	-1	0	+1	+2
STYLE-STIGMA	stigma papillae differentiate		the style elongates, the stigma changes from green to yellow		stigmatic exudate appears		the stigma turns brown	
COROLLA	green	becomes longer than the ovary		begins to turn orange- yellow	orange yellow	opens in the early morning and closes 6 h later		wilts
GERMINATED POLLEN GRAINS(%)					53±12	71±17	27±9	16±5
POLLEN TUBES AT THE BASE OF THE STIGMA					+	+	+	+
POLLEN TUBES AT THE BASE OF THE STYLE					+	+	+	
FERTILISATION					+	+		

Fig. 1. Characteristics of pistil parts of *Cucurbita pepo* in different stages of development before and after pollination. The germination percentage (mean±SD) of pollen on stigmas of different ages and the position of pollen tubes (+) are also reported.

association was evaluated by a Spearman's rank correlation coefficient.

RESULTS

The female flower before and after anthesis

Two days before anthesis, the petals began to turn yellow at the edges, becoming orange-yellow the day before anthesis (Fig. 1). Stigmatic exudate appeared the day before anthesis (Fig. 1) and became more abundant on the day of anthesis. The stigma began to turn brown the day after anthesis and this colour was increasingly evident from the second day onward. Female flowers were open from 5.30-6.00 am to 11.00-12.00 am during the study period (Nepi and Pacini 1993).

Growth of the ovary and corolla showed a similar trend until seven days before anthesis, when the corolla began to grow faster; corolla growth was accentuated in the last three days before anthesis (Fig. 2) when the style elongated at a greater rate and the stigma increased in volume and gradually changed from green to yellow (Fig. 1).

Fig. 2. Growth of corolla (■) and ovary (○) of *Cucurbita pepo* 10 days before anthesis to the day of anthesis. The two floral parts are approximately the same length until 7 days before anthesis, after which the growth of the corolla exceeds that of the pistil.

Pistil structure

Cucurbita pepo cv. Tonda di Nizza has an inferior round-ovary which contains about 400 ovules arranged in six rows, except at the top of the ovary where they are only three rows (Fig. 3). The stigma is trilobate of the multicellular, papillate, wet type (Heslop-Harrison and Shivanna 1977). Each lobe has two bundles of transmitting tissue that unite at the base of the stigma to form a single transmitting bundle (Fig. 3). The style therefore contains three closely apposed stilar transmitting bundles (Fig. 3A). In the receptacle region, the three bundles partially merge and decrease in size (Fig. 3B); at a distance of 0.5 cm from this junction, they increase in size and expand into two peripheral wings that define three locules (Fig. 3C). At the mid length of the ovary ovules are arranged in six rows (Fig. 3D). The wings terminate before the upper part of the ovary (Fig. 3C), whereas the lower part consists almost of wings only (Figs 3E and 3F).

Pistil receptivity

Irrespective of the method of investigation used, stigma receptivity was found to begin the day before anthesis and

Fig. 3. Diagram of the arrangement of the transmitting tissue of *Cucurbita pepo* from the stigma surface to the ovules. On the left, one of the three bundles is shown in three dimensions, on the right various transverse sections of the pistil. Each lobe of the stigma contains two bundles that unite at the base of the stigma, giving rise to a single bundle of transmitting tissue of even thickness that runs the whole length of the style (A). In the receptacle region, the bundles of transmitting tissue become considerably thinner (B) and expand again in the ovary, each forming two wings, and together demarcate three locules containing the ovules (C and D). The wings are not present in the upper part of the ovary (about 0.5 cm) (C) and the central axis of the transmitting tissue does not reach the base of the ovary (E and F).

to continue until two days afterwards. The stigma is therefore receptive for four days (Fig. 1). The pollen tubes were emitted from 3 to 5 minutes after pollination, irrespective of stigma receptivity. They grew along the papilla surface and penetrated at its base (Fig. 4). Two and three days before anthesis, the pollen adhered to the stigma but did not germinate. Since pollen viability is brief (Nepi and Pacini 1993), it died before the stigma became receptive. If pollination occurred on the day of anthesis or the previous day (Fig. 1), the tubes germinated, penetrated the transmitting tissue and fertilisation occurred. When pollination was carried out one day after anthesis, the tubes reached the upper part of the ovary but fertilisation did not occur. Two days after anthesis, the tubes penetrated the stigma but stopped growing before they reached the style (Fig. 1). In both the last two cases, the ovary grew slightly for 3-4 days after pollination (Fig. 5) and then began to degenerate.

Pollen tube emission and growth

The mean number of pollen grains remaining attached to the stigma after hand pollination on the day of anthesis was 917 ± 321 (mean $\pm$ SD). The percentage of pollen grains

Fig. 4. Stigmatic surface of *Cucurbita pepo* with pollen grains 15 min. after pollination. The pollen tube grows along the surface of the papilla and penetrates at its base. Bar = 40 μ m. Pt = pollen tube; sp = stigmatic papillae.

Fig. 5. Increase in diameter of fruit of *Cucurbita pepo*. Fruit diameter increases at a faster rate when the stigma is pollinated on the day of anthesis (□) than when pollination occurs on the day before anthesis (■). The increased rate of growth is already detectable two days after pollination. When pollination occurs one (●) or two (○) days after anthesis, the ovary grows at a slower rate in the next 3–4 days and then begins to degenerate.

that germinated on the stigma varied with pistil age: it was highest (71%) at anthesis, slightly lower (53%) before anthesis and much lower (27% and 17% respectively) one and two days after anthesis (Fig. 1).

In the stigma-style region, the tubes grew in the bundle corresponding to the stigma lobe on which the pollen had been deposited. When the pollen tubes reached the ovary, they were able to pass from one bundle to another.

The rate of pollen tube growth varied in the different regions of transmitting tissue and according to the period of receptivity in which pollination occurred (Fig. 6a, b). When pistils were pollinated one day before anthesis, the tube grew very fast in the stigma-style region, reaching the distal extremity in only two hours (Fig. 6a) with a mean velocity of 6 mm/h (Fig. 6b). The rate of growth decreased considerably when the tubes reached the ovary (Fig. 6b), they were in the middle part of the ovary at 28 hours after pollination and continued to grow in the successive hours (not reported in Fig. 6a, b). In pistils pollinated at anthesis, the tubes grew slowly in the stigma-style region with a mean velocity of 3 mm/h, but in the ovary they grew faster than they had on the previous day (Fig. 6b). When pollination was carried out the day after anthesis, the tubes grew at an intermediate speed in the stigma-style section, but did not go beyond the upper part of the ovary (Fig. 6b). When pollination occurred two days after anthesis the tubes penetrated the first millimetres of the stigma in the first two hours but did not grow anymore in the following hours (Fig. 6a).

Fruit growth and seed set

For pollination at anthesis or the day before, the growth curves of the fruits were sigmoidal and growth was greatest between day 2 and day 12 (Fig. 5). Fruit diameter increased faster after pollination on the day of anthesis than on the previous day (Fig. 5). In both cases maximum size was reached about 16 days after pollination (Fig. 5). There are significant differences in the final fruit weight according to the time of pollination. When pollination occurred at anthesis, mean fruit weight was greater (1128.76 g) than for fruits developing as a result of pollination on the day before anthesis (941.56 g) ($t = -6.41, p < 0.05$) (Fig. 7a, b).

Fig. 6. Pistil receptivity and pollen tube growth in *Cucurbita pepo*. A. Distances reached by pollen tubes in pistils of different ages. The pollen tubes penetrated the stigma, grew in the transmitting tissue of the style and fertilised ovules only if pollination occurred at anthesis (white column) or the day before anthesis (hatched column). When pollination was carried out the day after anthesis (dotted column) the tubes reached the upper part of the ovary but did not fertilise the ovules. Two days after anthesis (black column) the tubes penetrated the stigma but did not continued to grow in the bundles of transmitting tissue. B. Pollen tube growth rates in pistils of different ages. The fastest growth was recorded on the day before anthesis in the first two hours after pollination, however these pollen tubes grew only slowly into the ovary. On the day of anthesis the tubes grew more slowly in the style-stigma section but faster into the ovary.

The mean number of seeds per fruit was greater when pollination was performed at anthesis (281.56) than when it was performed the day before anthesis (171.98) ($\alpha = 0.05, t = -10.69, p = 0$) (Fig. 8a, b). There was a positive correlation between seed number and fresh weight of the fruit when pollination was carried out the day of anthesis ($r = 0.75, \alpha < 0.05$) (Fig. 9a) and the day before ($r = 0.76, \alpha < 0.05$) (Fig. 9b).

DISCUSSION

Female receptivity

Female receptivity varies widely in angiosperms (Richards 1986). Generally speaking, wind pollinated plants have a longer period of female receptivity than insect pollinated plants (Khadari et al. 1995). Among the latter, however, there are plants with long female receptivity, such as certain orchids (Endress 1994) and some species of the genus *Ficus* in which syconium receptivity is regulated by insect activity and ceases soon after the pollinator has done its work (Khadari et al. 1995).

Pistil receptivity in *Cucurbita pepo* comprises stigmatic, stylar and ovular receptivity. Pollen tubes do not grow into

Fig. 7. Normal fitted distribution curve of fruit fresh weight when the pistil was pollinated at anthesis (a) and on the day before (b).

the ovary two days after pollination. This fact could be due to a short ovule receptivity or to an incipient degeneration of the transmitting tissue that supports pollen tube growth. Styler receptivity lasts three days, from the day before to the day after anthesis. Stigma receptivity lasts four days, from one day before to two days after anthesis. In kiwifruit, the EPP is limited by short stigma receptivity (Gonzales et al. 1995). It is interesting that in *Cucurbita pepo* the pollen adhered to the stigma but only germinated in the presence of stigmatic exudate. Although pollenkit is very abundant in *Cucurbita pepo* (Ciampolini et al. 1993) and necessary for adhesion, it is presumably not sufficient to cause germination. Pollen did not germinate on the stigmatic papillae until the exudate appeared the day before anthesis. The period of stigma receptivity also influenced the pollen germination percentage which was much higher during the EPP than on the other three days in which the pollen succeeded in germinating on the stigma.

As far as seed set is concerned, our results showed that the female flower was receptive one day before anthesis.

Fig. 8. Normal fitted distribution of number of seeds per fruit when the pistil was pollinated at anthesis (a) and the day before (b).

However in nature, pollination is limited by the short, only six hours, period of anthesis.

Pollen tube growth

In most plants, including *Cucurbita pepo*, pollen tube growth lasts 12 to 48 h, from pollen germination to fertilisation, in the case of pollination at anthesis (Richards 1986). Pollen tubes may grow faster than 2 mm/h (Pierson and Cresti 1992) and their growth can be regarded as an example of high secretory activity. Growth rates vary widely and depend on many factors, including pollen vigour (Shivanna et al. 1991) and temperature (Richards 1986). Differences in the vigour of pollen grains of the same species are the consequence of different pollen genotypes and lead to differences in the vigour of the progeny (Mulcahy 1979). Pollen tube growth is totally inhibited above 40°C and below 10°C in many species (Richards 1986). Winsor and Stephenson (1995) reported a rather constant growth rate of about 30 µm/h in the first 22 h for the mass of pollen tubes in flowers of *Cucurbita pepo* cv

Fig. 9. Correlation between fruit fresh weight and number of seeds per fruit when the pistil was pollinated the day of anthesis ($r=0.75$, $\alpha<0.05$) (a) and the day before ($r=0.76$, $\alpha<0.05$) (b).

Black Beauty Bush pollinated at anthesis. In the same cultivar, Stephenson et al. (1988b) reported that most pollen tubes reached the middle part of the ovary within 24 h of pollination, when this occurred at anthesis. Our observations agree with those of Stephenson et al. (1988b), despite the fact that we used the cultivar Tonda di Nizza.

The present results show that pollen tube growth rate is not the same in all parts of the pistil and that it also depends on female receptivity. As observed by Winsor and Stephenson (1995) in *Cucurbita pepo* cv Black Beauty Bush, pollen tube competition is not the same at all points along the path from the stigma to the micropyle and pollen attrition may vary in relation to the thickness of the transmitting tissue of the style. The conformation of these transmitting bundles, which are independent but join partially in the receptacle region, suggests that some tubes may avoid high competition in a bundle by escaping into a contiguous bundle in which competition may be less.

The highest pollen tube growth rate was observed in pistils pollinated one day before anthesis. This is probably due to the fact that all the biochemical mechanisms of recognition of pollen tubes in the style-stigma section are not yet fully active, and is the reason why some incompatibility barriers can be overcome by bud pollination (van der Knaap and de Ruiter 1978).

The pollen tube growth rate of *Cucurbita pepo* was high compared with that reported for other Cucurbitaceae such as *Luffa cylindrica* and *Cucumis melo*, at least in the first hours after germination. However in these species the growth rate was measured in vitro. In *C. melo* the pollen tubes have a mean length of 680 µm after an hour of incubation (Cuny and Roudot 1991). The tubes of *L. cylindrica* reach a mean length of about 500 µm after 3 h of incubation (Setia et al. 1990). The high growth rate is probably also due to the fact that the pollen of *Cucurbita pepo* at the moment of dispersal has a low grade of dehydration (is slightly dehydrated) and to the presence of pre-formed callose either in the sporoderm and in cytoplasmic vesicles (Nepi et al. 1995). *Luffa cylindrica* and *Cucumis melo*, on the other hand, have pollen with a higher grade of dehydration (partially dehydrated) at dispersal and takes more time to rehydrate before it can emit pollen tube (our unpublished data).

Pollen tube path and competition

In angiosperms, competition between male gametophytes can occur from the time of deposition on the stigma until fertilisation occurs. With entomophilous pollination, a surplus of pollen with respect to the number of ovules to be fertilised may be deposited on the stigma and this provides the potential for gametophytic competition and selection (Murdy and Brown Carter 1987). The number of tubes reaching the ovules may be reduced in two main ways: by differences in pollen competitive ability and by modulation of these differences by the pistil (Hormaza and Herrero 1996). This modulation also includes physical constraints, such as a reduction in the thickness of the transmitting tissue (Herrero 1992; Hormaza and Herrero 1996). Winsor and Stephenson (1995) noted that in *Cucurbita pepo* cv Black Beauty nearly all the pollen contacting the stigma germinated, suggesting that most attrition occurs during the stage of pollen tube growth. In *Cucurbita pepo* cv Tonda di Nizza the transmitting tissue constricts in the receptacle

region; the fact that pollen tube growth rate decreases at the lower end of the style suggests that pollen tube competition is enhanced in the receptacle region. The pollen tubes can only pass from one bundle of transmitting tissue to another when they reach the ovary. The width of the transmitting tissue is much greater in the ovary and this presumably causes low competition between pollen tubes, also because the number of ovules to be fertilised is large. We can conclude that competition among pollen tube is essentially completed prior to the pollen tubes entering the ovary.

The capacity of the pollen tubes to pass from one bundle of transmitting tissue to another may decrease the frequency of morphological anomalies in the fruits due to heterogeneous distribution of pollen on the three stigmatic lobes. This capacity is of commercial value because it guarantees a quality product in terms of shape even when only one or two stigmatic lobe has received good pollen load. An anatomical structure is involved in distributing pollen tubes evenly around the ovary in kiwifruit (Howpage et al. 1998), whilst in maize the pollination of a sector of the silk led to fertilisation of only that sector of the cob (Heslop-Harrison et al. 1985).

Since the pollen germinates only a few minutes after pollination and since pollinators bring 827 ± 143 pollen grains in 78 ± 16 visits (mean $\pm$ SD) during the six hours of anthesis (Nepi and Pacini 1993), male gametophytic competition should not be very high. On the other hand, competition between ovules has been documented (Stephenson et al. 1988b).

Fruit maturation and seed set

It is commonly accepted that a positive relationship, albeit not universal, exists between pollination intensity, seed number and fruit maturation (Lee 1988). Grape fruits with fewer seeds tend to grow faster, although the final volume is less than that of fruits with more seeds (Staudt et al. 1986). According to Stephenson et al. (1985, 1988a, b), fruit of *Cucurbita pepo* cv Black Beauty derived from a high pollen load grows faster, reaches a larger size and contains more seeds than fruits derived from a low pollen load. The results of this study support the positive relationship between fruit size and number of seeds.

Our data show that the period of female receptivity in which pollination occurs may also influence fruit maturation and seed set in *Cucurbita pepo*. This influence has also been found in *Cucumis sativus* (Le Deunff et al. 1993) and *Trifolium repens* (Jacobsen and Martens 1994). In *Cucurbita pepo* cv Tonda di Nizza, the fruits grew bigger and had more seeds when pollination occurred at anthesis. It has been demonstrated that fruits with few seeds are more likely to abort (Stephenson 1988a; Lee 1988). Few seeds in a fruit may be due to low pollen load or to the position of the fruit on the plant or in the inflorescence (Lee 1988). In many species, fruits located closest to the source of nutrients are more likely to mature than others. We can also add that because it affects seed number, female receptivity may also influence abortion of fruits.

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WPLYW WIEKU SŁUPKA NA WZROST ŁAGIEWKI PYŁKOWEJ,
ROZWÓJ OWOCÓW I ZAWIĄZYWANIE NASION U *CUCURBITA PEPO* L.

STRESZCZENIE

Badano wpływ wieku słupka na wzrost łagiewki pyłkowej, rozwój owoców i zawiązywanie nasion u *Cucurbita pepo* L., której kwiat otwiera się tylko na okres sześciu godzin. Wrażliwość znamienia słupka trwa cztery dni, począwszy od dnia poprzedzającego do dwóch dni po pełnym rozkwicie. Wrażliwość zalążka trwa dwa dni, w dniu pełnego rozkwitu i w dniu poprzedzającym pełny rozkwit. Tempo wzrostu łagiewki pyłkowej w poszczególnych częściach słupka różni się w zależności od wieku. W okolicy znamienia i szyjki słupka, łagiewki rozwijają się szybciej jeśli zapylenie następuje w dniu poprzedzającym zakwit; w zalążni rosną one szybciej jeśli zapylenie następuje w czasie zakwit. W okolicy dna kwiatowego, gdzie tkanka przewodząca jest zredukowana, tempo wzrostu jest mniejsze niezależnie od czasu zapylenia. Owoce są większe i cięższe jeśli zapylenie następuje w fazie kwitnienia. Stwierdzono dodatnią korelację pomiędzy liczbą nasion i wagą owoców, jeśli zapylenie nastąpiło w fazie kwitnienia i w dniu poprzedzającym.

SŁOWA KLUCZOWE: *Cucurbita pepo*, wiek słupka, wzrost łagiewki pyłkowej, konkurencja łagiewki pyłkowej, zawiązywanie nasion, zawiązywanie owoców.