



**ANALYSIS OF MORPHO-PHYSIOLOGICAL DIVERSITY OF SOME TUNISIAN DATE PALM POLLINATORS AND STUDY OF THEIR WHORL POLLEN GRAIN ON FRUIT SET, THE TIME OF RIPENING AND YIELD OF DATE PALM CV. DEGLET NOUR**

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| ARTICLE INFO  | ABSTRACT   |
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| Received 16 <sup>th</sup> February, 2016<br>Received in revised form 27 <sup>th</sup> March, 2016<br>Accepted 15 <sup>th</sup> April, 2016<br>Published online 28 <sup>th</sup> May, 2016 | In Tunisia, the date palm is characterized by considerable genetic richness represented by 250 cultivars. It represents the backbone of the agricultural activity in the south west of the country. In this work we have done in the first step a study of morpho-physiological diversity of 10 date palm pollinator selected and on second step the effect of pollen on to maturity and fruit quality of the variety 'Deglet Nour'. the principal component analysis (PCA) using the R software allowed to choose two axes: axis 1 (F1) and the axis2 (F2) that absorbed a maximum of existing variability between pollinators date palm studied (48.80%). Their values are 28.79% and 20.01% respectively. The result showed that only 16 characters from 51 traits tested, can be used as a discriminative character. So pollinators showed a significant discrepancy between them, with a similarity index ranging from 0.174 to 0.764 that can be grouped into 4 groups. For the metaxenic effect of pollinators on the date of ripening and fruit quality, The analysis of the effect of different pollinators of fruit ripening has identified three groups: Pollinators hastening fruit ripening: P121 and P45 Pollinators retarding fruit ripening: P invitro and Pollinators have an effect on unstable maturation: remaining pollinators, the obtained result showed that the P121 and is the better pollinator because it promotes the maturation and gives a good caliber. The maximum fruit yield was scored with P Invitro. |
| <b>Keywords:</b><br>Pollinators, Genetic Diversity, Morphological Marker, Fruit Set, Yield.   |  |

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**INTRODUCTION**

The date palm, *Phoenix dactylifera* L., is probably the oldest cultivated plant culture by man on earth. This is an iconic desert plant, which is cultivated since ancient times in arid and semi-arid regions of the planet. Its fruits have a very high energy food and commercial value. This perennial monocot species also has an ecological importance capital by creating in the desert, a favorable microclimate for the development of the underlying cultures. In Tunisia, the date palm plays a strategic role in the socio-economic stability of the oasis agro-ecosystem. Indeed, it is the main focus of agriculture in desert regions and provides the main financial resource of the oasis.

The Tunisian population living at the expense of phœniculture is estimated at about 10% [1]. World production of fruit from the tree of the date palm (*Phoenix dactylifera* L.) was estimated at 7.75 million tons with a value of 3.82 billion US dollars in 2012. Recall that Tunisia holds 25% of sales worldwide, and ranks fifth country in the world the harvest standpoint since Tunisia produces 9% of world output in dates [2]. Nationally, total production of dates positively changes over time, because these statistics predict a date harvest of about 246,000 for 2015 tons against 223,000 tons in the previous season [2], of which 72% of the variety of "Deglet Nour". A very important part of this variety is exported it reaches 95466 tons in 2013, representing 4% of the overall value of agricultural production

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and 13% of the total value of agricultural exports. Besides, exports of dates occupy the 2nd place among the agri-food exports after olive oil [2]. It developed as an active industrial and commercial sector, export oriented. Date palm production in quantity and quality is closely linked to the pollinators 'dokkars' the fact that the quality of the pollen grain is a key factor of performance. DARLEEN *et al.* 1988 mentions that the pollen influences not only the size and shape of the seeds (xenia), but also the size, shape, weight and fruit maturation rate (metaxenia). Therefore, it appears that the pollen plays a significant role in the expression of certain characteristics of the fruit. It is therefore necessary to conduct studies to detect any differences between the different types of pollen in order to compile a list of 'dokkars' with high quality of pollen.

However, Surveys and research for many years throughout the national territory have been directed mainly towards the female palms, and little research on the male palm (Dokkars). In this context, a number of works were carried out between 1985 and 1994 by Dr. Larbi BOUGHEDIRI (Professor at the University of Annaba) on pollens from the region of Biskra and contributed to a better understanding of the criteria that could to characterize the quality of the pollen of date and their use in distinguishing between different males palms. It is in the same context, we present this preliminary search on pollen male feet from the Tozeur region. Our Work are composed on two parts conducted according to the following steps, first we analysed the genetic diversity of pollinators by physiological and morphological markers. Second we study the metaxenic effect of these pollinators on the duration of ripening dates of the variety Deglet- Noure and the quality of the obtained date.

## MATERIALS AND METHODS

### *Plant and Experimental Sites*

The selected sampling sites are located in the region of Tozeur (male's collection) and Dgueche (plot of CRRAO) and it were used for the morphological characterization. Cross-pollination was carried out in two different sites one is Deguehe (plot of CRRAO) and the other in Tamarza (private plot). The study was performed on a collection of 10 date palm pollinators, they were coded with the letter P followed by a number or a name: P76, P176, P96, P45, P30, P121, P8, P7, P sami and P invitro, including pollinators P8, P7, and P sami P invitro are obtained from the experimental plot of the Regional Research Centre of Oasis Agriculture (CRRAO) of Degueche and P76 pollinators, P176, P96, P45, P30, P121 are in the plot of CRRAO in Tozeur. As for female feets; "Deglet Nour" variety was chosen to perform cross-pollination, two of which are in the plot of Deguech and the other were in a Mides mountain oasis in the region of Tamarza.

### *Morphological Study*

Morphological studies are determined using the date palm tree descriptor developed by IPGRI in 2005 [3] (International Plant Genetic Resources Institute). This study aims to establish a fact sheet and to determine the genetic relationships between different types of pollinators through morphological markers.

### *Descriptors Growth*

This parameter includes sex; vigor ; the port; the appearance of the crown; the shape of the stipe; persistent of cornaf; the presence of air emissions; fluff mane presence and ability to produce suckers.

### *Descriptors of Palms*

In the description of palms, several parameters are measured beings namely the level of curvature of the palm; the rotation of the palm; the total length of the fin (m); the total width of the fin (m); the maximum length of the spinous portion; the thickness of the spine; the color of the petiole; the width of the palm-based petiole (cm); the average number of thorns; the rigidity of thorns; the number of needles per cluster type; the maximum thickness of the spine of the middle (cm); the maximum length of the spine of the medium (mm); the maximum length of the spine (cm); by the number of quills palms; color penne; flexibility penne; the penne group; apical divergence penne; the maximum width of penne amidst the palm (cm); the maximum length of the quill in the middle of the palm (cm); the length of the apical quill (cm); and the width of the apical quill (cm).

### *Inflorescences Descriptors*

This parameter has the form of husk, the total number of husks, the speed of development, the total length of the husk (cm), the width of the husk (cm), the weight of husk (Kg), density spikelet, number of spikelets per bunch, the shape of the spikelet, the length of the spikelet longest (cm), the length of the spikelet shortest (cm), the length of the part with flower (cm) and the number of flowers of the two longest and shortest spikelets.

### *Pollen Descriptors*

This parameter includes the weight of pollen (g), its productivity, its smell and its color.

### *Physiological Study*

#### *Pollen Harvest*

Harvesting of inflorescences was made just burst husk to avoid mixing pollen. Then pollen grains were released and left to dry in the open air for two days. After, spikelets were cut to their insertion point using a chisel and then spread them on kraft paper and placed in a ventilated area, away from drafts and the humidity and ambient temperature .The monitoring of spikelets is done daily to prevent mold growth. The spikelets are then picked up and placed in a sieve with a lower porosity than 1 mm, the pollen grains are then placed in glass dishes and sealed.

### *Sustainability Test*

This is a simple color test that estimates the quality of pollen based on the calculation of the percentage of viability (colored pollen compared to unstained pollen). We used Acetocarmine as cytoplasmic dye. This dye has the ability to intercalate into the core to give it a red coloring. Viable pollen stain red and acquire a spherical shape. Non-viable pollen does not stain and have a wrinkled appearance. The viability rate is calculated using the following formula: % viability = (Number of viable grain) / (Number of total grains)

### **Study of the Metaxenic Effect Pollinators on the Date of Ripening and Fruit Quality Cross-Pollination**

For realizing this experimentation three females date palm tree of 'Deglet Nour' variety were used, two palms are at Degueche and one at Tamarza, on each palm tree we have chosen three flower stalks as three repetition. The technique is used for the first time by Nixon *et al.* 1928 [6] and by recovery Lakhoua *et al.* 1966 [7] and Ben Abdallah *et al.* 1986 [8]. It is bagging the inflorescence before bursting of the spathe (kraft paper bag) and monitor it through each day to remove the husk from its bursting. The spikelets of the same system, thus released, are divided into ten groups of five spikelets, each group is isolated in a sleeve of kraft paper, the sleeve is held in place by a string at the base of the strands and the other end folded being closed by a double fold held by a clip. Pollination was carried out by removing the paperclip in this brochure end of the bag and introducing three male spikelets in the diet group and each group will be marked with a label on which is labeled the name of the pollinator used. Special care was taken by passing their hands with alcohol before the operation and during the passage of a pollen to another. The bags are removed one month after pollination, only circling spikelets groups labels still pending.

### **The Study of Metaxenic Effect of Pollinators on the Date of Maturity**

To determine this effect we have studied the evolution of fruit from the 'Bissir' stage 'Tamar' stage. Indeed, when the first fruits reach the stage 'BISR', characterized by the color change from green to yellow, the counting is done weekly and the percentage of 'Bissir' is noted. At the entrance to the 'Tamar' stage, distinguished by the color change from yellow to brown fruit, percentages of 'Bissir', 'Tamar' and parthenocarpic fruits (without core) are determined using the following formulas:

- % Bissir =  $100 \times B / (FT-P)$
- % Tamar =  $100 \times T / (FT-P)$

B = Number of fruits have reached the stage 'Bissir' on a group of five ears.

T = Number of fruits have reached the stage 'Tamar' on a group of five ears.

P = Number of parthenocarpic fruit on a group of five ears.

FT = B + T + P = Total number of fruit on a group of five ears.

### **Study of the Effect of Pollen on the Quality and Yield of Dates**

During harvest, each specific group of pollen is deposited in packaging bearing the reference group (palm, flower stalk and pollinator number). Three fruits of each group are taken randomly and measured using a caliper. The length and width of the fruit as well as the core as well as the core and Fruit Pulp are Weighted

### **Statistical Analysis of Data**

#### **Principal Component Analysis (PCA)**

This is an effective method for the analysis of quantitative data, based on the correlations. It allows to visualize and analyze correlations between variables and the dispersion in the data.

This method allows you to set markers that contribute to the description of the variability and obtaining a graph or presentation projection of individuals according to their similarity in terms of the axes of the ACP [9]. The CPA in this study was performed using the software R (2014).

#### **The Genetic Similarities**

Morphological data were submitted to SIMQUAL program (similarity for qualitative data program) of NTSYS-pc software (the numerical taxonomy and multivariate analysis system for personal computer) version 1.7 [10] to generate the matrix of similarities between pollinators. The similarity coefficient were used for analysis program under WPGMA "weighted pair-group method using Arithmetic" of NTSYS software to establish the dendrogram corresponding to the grouping of different studied pollinators [10].

## **RESULTS AND DISCUSSIONS**

### **Morphological Parameters**

Morphological characters are considered by farmer for the choice of pollinators of date palm, based primarily on the quality of the inflorescence. Measured growth parameters are quite variable among the pollinators. The pollinators p8, p76 and p176 are characterized by a high ability to produce suckers. Indeed, this character is interesting because it allows the multiplication of these pollinators because this kind of multiplication is the mode of reproduction that preserves the morphological characters of mother plants.

### **Principal Component Analysis**

Morphological parameters are processed by a principal component analysis (PCA) using software R (2014). We chose axis 1 (F1) and axis 2 (F2) which absorbed a maximum of existing variability between pollinators date palm studied (48.80%). Their values are 28.79 % and 20.01 % respectively. (Figure 1). According to the result obtained with the R software, the axis 1 represents 28.79% of the variability. It is positively defined by this parameters: the maximum length of the spine (C24), the width of the palm at the base of petiole (C19), the total length (C13), the maximum width of the husk (C38), the length of the spikelet longest (C43), the length of the spikelet shortest (C44) Number of spines by grouping type (C41) and finally the average number of thorns (C20). It is concluded that these parameters are most discriminating with respect to this axis. For axis 2 that absorbs 20.01% of the variability, it is positively defined by this parameters: the maximum width of the apical penne (C33), the length of the apical penne (C32) and the weight of the spathe (C39) and negatively by a single parameter which is the maximum width of the fin (C14). The projection in the plane defined by the two axes was shown in the figure 1. The analysis of this figure shows that there are five distinct groups: The first group A only carries the P30 pollinator and P121 that are highly correlated with F2, so these two different pollinators other by the characters defining this axis which are the maximum width of the apical penne, length of apical penne, weight of the husk and the maximum width of the fin. The second group B is represented by P45 and P176 pollinators that correlate negatively with F1, so they gather by

the defining characters and discriminate it from others by the same characters. The third group C consists of a single pollinator P invitro which is negatively correlated to both axes. Next, the group D is composed of a single pollinator P sami which is negatively correlated to both axes and finally the group E carries the pollinators P8, P7, P96 and P76, which are positively correlated with the F1 axis.

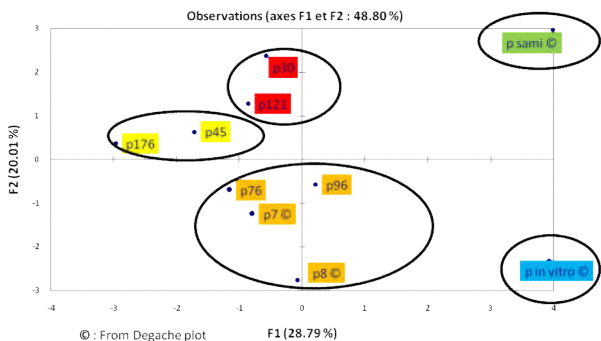


Figure 1 Dispersion of Ten Pollinators on the Plane Defined by the First two Components of the PCA

**Genetic Similarity**

The resulting matrix (Table 1) disclosed similarity coefficients ranging from 0.174 and 0.764. These values express divergent groups at the plant material studied. Thus, it is found that the couple "P176" - "P96", with 0.174 coefficient is largely distant. Its divergence lies substantially at the morphology of the fin through the total length (C13), the maximum width of the fin (C14), the thickness of the spine (C17), the color of the petiole (C18), the width of the palm at the base of the petiole. Similarly, these pollinators are distinguished by the color (C50) and the weight of pollen (C51). For against, the highest similarity (0.764) was observed between the couple of pollinators "P121" - "P30", which are similar in a larger number of characters as the maximum width of the apical quill (C33), the length of apical penne (C32), the weight of the spathe (C39) and the maximum width of the palm (C14).

Table 1 Genetic Similarity Matrix SM Based on Morphological Markers Established by the NTSYS-pc Program Version 2.02

| P8   | PIV       | PSA       | P45       | P176      | P176      | P121      | P30       | P96       | P76       |           |
|------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|
| p8   | 1.0000000 |           |           |           |           |           |           |           |           |           |
| PIV  | 0.3333333 | 1.0000000 |           |           |           |           |           |           |           |           |
| PSA  | 0.3333333 | 0.3725490 | 1.0000000 |           |           |           |           |           |           |           |
| P7   | 0.4509804 | 0.3333333 | 0.3529412 | 1.0000000 |           |           |           |           |           |           |
| P45  | 0.2941176 | 0.2745098 | 0.2549020 | 0.2549020 | 1.0000000 |           |           |           |           |           |
| P176 | 0.3725490 | 0.2549020 | 0.2549020 | 0.2941176 | 0.2745098 | 1.0000000 |           |           |           |           |
| P121 | 0.3725490 | 0.3529412 | 0.2941176 | 0.4117647 | 0.2549020 | 0.3529412 | 1.0000000 |           |           |           |
| P30  | 0.3529412 | 0.3529412 | 0.2941176 | 0.3921569 | 0.2352941 | 0.3529412 | 0.7647059 | 1.0000000 |           |           |
| P96  | 0.3137255 | 0.4117647 | 0.3137255 | 0.3137255 | 0.1764706 | 0.2745098 | 0.4117647 | 0.4117647 | 1.0000000 |           |
| P76  | 0.3333333 | 0.2156863 | 0.2745098 | 0.3333333 | 0.2549020 | 0.3725490 | 0.2352941 | 0.2156863 | 0.2156863 | 1.0000000 |

**Genetic Diversity**

The matrix generated by the different morphological parameters analyzed by the WPGMA analysis using the NTSYS software has established the genetic relationships between pollinators. Thus the dendrogram obtained revealed the existence of phenotypic polymorphisms. For the ten studied pollinators, three groups were distinguished (Figure 2). The first group A comprises only P45 pollinator. This is

characterized by the highest maximum thickness of the spine (6.3mm). The second group B is represented by two P76 and P176 pollinators whose common characteristic is the ability to produce high emissions. The third group C is divided into four sub groups: Overall C-1 is only represented by the pollinator Psami. This pollinator is characterized by the highest maximum length of the spine (13.2 cm). C-2 is represented by two P96 and Pinvitro pollinators. Indeed these pollinator have several common characteristics such as the maximum thickness of the spine (5 mm), flexibility penne, density of spikelets. The third subgroup is represented by two pollinators P30 and P45 and the last is composed of the two pollinators P7 C et P8 C. This grouping more or less confirms the results obtained by analyzing multi ACP varied.

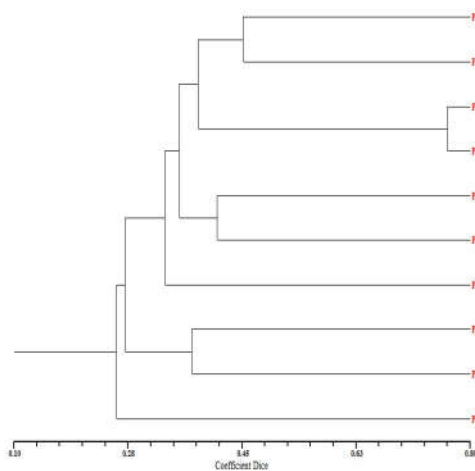


Figure 2 Dendrogram Grouping Pollinators Prepared According to WPGM Method Based on Morphological Markers

Morphological studies of date palms were still considered difficult to implement since they require a large set of phenotypic data and because the date palms are quite varied because of the environmental effect [12].

In this study we attempted to set up an experiment based on the use of tools for morphological characterization of pollinator groups and we identified those most discriminating. Therefore, we assume that the feet studied are characterized by a fairly high level of genetic diversity. This is strongly supported by projection of pollinating in the main plane of the PCA and also in WPGMA cluster analysis. The most discriminating characteristics according to our principal components analysis

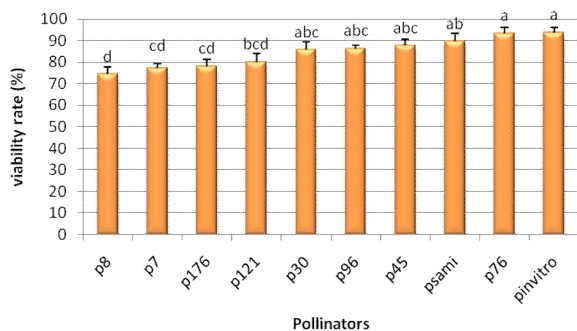
are the maximum length of the spine (C24), the width of the palm at the base of the petiole (C19), the total length (C13), maximum width the spathe (C38), the length of the spikelet longest (C43), the length of the spikelet shortest (C44), the number of thorns by type of grouping (C41), the average number of Thorns (C20), the maximum width of the apical penne (C33), the length of the apical penne (C32), the weight of the spathe (C39), maximum of palm width (C14), the total length of the spathe (C37), the maximum length of the feathers in the middle of the palm (C31), the thickness of the spine (C17), maximum width penne amidst the palm (C30). Our data generally agree with those reported for the 11 cultivars and the male foot Mauritanian studied by date palm [13] based on 18 phenotypic markers.

The analysis resulted in the main component 12 discriminating characteristics that are the width of the sheet, the length of petiole of the spine thickness, the number of pinions, the length of the fin, the length of the spinous part, the length the penne in the middle of the palm, the ventral angle of the middle penne, penne the width of the middle of the palm, the apical angle of the palm, length of apical penne, penne width of the .Quant apical to the physiological study, our results are consistent with those found by [14] who consider the viability of the pollen levels in the date palm is between 40 and 100%. These results are also consistent with those of [15], who found that the rate of non-viable seeds does not exceed 30% for the date palm. Thus, the study of polymorphism in this collection pollinator through phenotypic markers is essential.

**Physiological parameters**

**Viability of pollen grains**

The viability test was performed on fresh pollen. The figure 3 shows the different percentages of the ten pollinating pollen viability. The statistical analysis of this variable showed significant differences in rates of pollen viabilities. The highest rate was obtained with the pollinator P invitro (93.78%) followed by pollinator P76 (93.40%) and the lowest was observed in the pollinator P8 (74.77%).



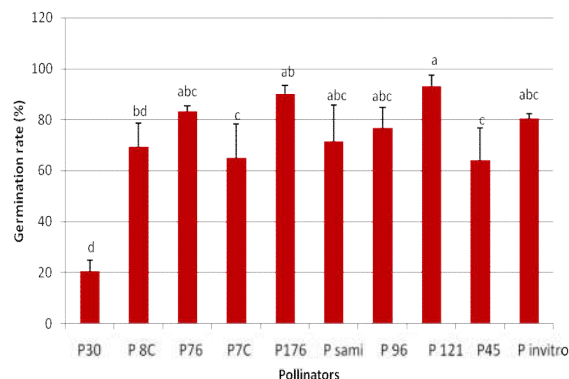
**Figure 3** Viability Rate of Pollinators

The values are statistically different by the test of Newman Keuls which explains the influence of the variety of pollinators on the viability rate. Thus, these results demonstrate that the viability of pollen grains is a criterion for distinguishing between different pollinators. Many investigation determined the pollen viability in some fruit species using the acetocarmine method such as [16,17], In addition to that Ream

and Furr [18] have reported that the rate of fruit set was closely related to percentage of viable pollen.

**Germination Rate**

Several media were tested in order to have the closest artificial environment of the real environment that is the stigma. For example Soliman *et al* [19], cited by Djerbi [20], evaluated a solid culture medium consisting of 1% agar and 2% glucose for 24 hours at temperature 27 ° C. Also, there was obtained a good germination on a solid medium composed of glucose and sucrose according to [14]. Germination provides information on the quality of pollen. The differences observed between the studied pollinators germination rate are statistically significant. The highest rates were observed among pollinators P121 (93.13%) and P176 (90.15%). the lowest was observed in the pollinator p30. The germination rate is significant effect on the morphological diversity. The average values of histograms topped by different letters are significantly different (Figure 4).



**Figure 4** Pollen Germination Rate of 10 Studied Pollinators

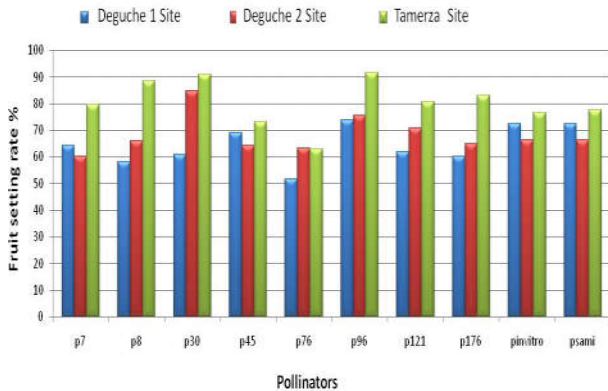
A pollen grain was considered germinated when issuing a pollen tube at least equal to its diameter. Germination information on the quality of pollen so it may be a basic criterion for the comparison of ten pollinators especially when the test applied Newman Keuls showed a difference between the percentages. More than 50% germination of pollen grains in laboratory ensures a high rate of fruit set. [21]. While Soliman *et al* [19], considers that pollen must germinate over 60% for a good fruit set rate in vivo. So P30 will not ensure a high seed set rate since it has a germination rate of 20%.

The reaction of pollen grains in the coloring includes evaluating the red color intensity after the cytoplasmic staining. This indicates the importance of the enzymatic activity and therefore pollen viability [20]. The results of the germination rate of these pollinators are not compatible with their viability rate. Indeed, P 30 has a low germination rate by 20% while its viability rate was 87% while P45 with a germination rate of 90% its viability rate has not exceeded 60% which is low among all pollinators. So we can conclude that the viability rate is not proportional relationship with the germination rate and any viable cell is not necessarily able to germinate [22]. This variation suggests the presence of genetic differences [19,21].

**Study of the Metaxenia Effect of the Pollen**

**Effect of Pollen on the Rate of Fruit Set**

The percentage of fruit set is an indicator of compatibility between pollinator and variety [22,23]. To compare the influence of different pollinators studied on female palms, we tried to compare the fruit setting rate obtained with each pollinator on the variety Deglet Nour. Figure 5 illustrates the effects of ten pollinator percentages of fruit set in the Degueche region with two repetition (palm tree 1 and palm tree 2) and the Tamarza region (palm 1). The differences are significant (Figure 5).



**Figure 5** Variation o Fruit Setting Rate According to Pollinators

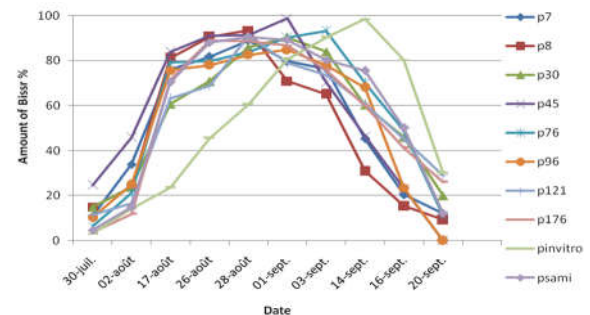
On Tozeur region and for the first palm tree, the highest fruit set rate is obtained by the group pollinated by P96 (73.86%) followed by Pinvitro and Psami with rates (72.59%) and (72.59%) while the lowest rate was obtained by the group pollinated by P76 (51.77%). With regard to the 2nd palm tree the highest fruit setting rate is obtained by the group pollinated by P30 (84.79%) followed by P96 and P121 with respective rate (75.67%) and (70.65%), while the most rate low was obtained by the group pollinated by P7 (60.05%).

As for the Tamarza region the highest fruit set rate is obtained by the group pollinated by P96 (91.59%) followed by P30 and P176 with rates (90.87%) and (83.26%) while the lowest rate was obtained by the group pollinated by P176 (83.26%). We can see that all pollinators can be used for pollination of the variety Deglet Nour and the fruit setting rate varies depending on pollinators, and finally foot chooses regions. We note also that the palm tree of the site Tamerza have the maximum attachment rate, compared to other feet, this can be explained by the effect of climate and environmental conditions.

Indeed, the site of Tozeur is characterized by its mode of modern culture with feet that are remote from one another. So it is more exposed to the wind blows in the spring that will increase the losses in flower spikelets, while the female foot Tamarza is located in an ancient oasis characterized by planting density and shading, which the wind is weak, so we can say that during this time, the wind is the main factor influencing the setting of flowers. these results are similar to those of Jalele *et al.* 2006 [23], Ibrahim and Shahid [24] and Iqbal *et al.* 2011 [25].

**Evolution of Fruit to "Bisser" Step**

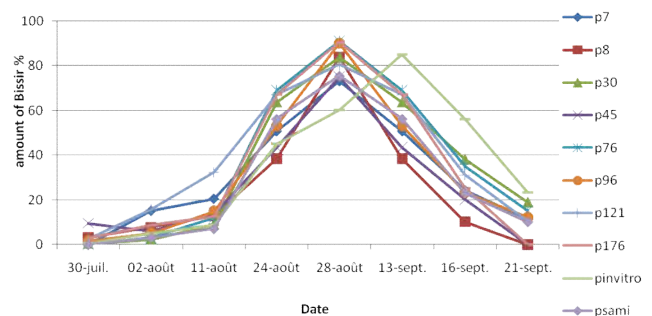
The fruits evaluation velocity at "Bisser" varies depending on the site and pollinators and also by the accumulated temperatures for a given period [22,24]. Indeed, in the Degueche region, and for the first data palm the amount of "Bisser" data contained in the pollinated groups P invitro reaches its maximum at the beginning of September while other pollinators except P45 which start from the beginning of August with a maximum amount of "Bisser" on the half of August (figure 6).



**Figure 6** Evolution of the Percentage of the Fruit at the Stage "Bisser" According to Pollinators in the Region Degueche 'Palm Tree 1

From first September the amount of "Bisser" starts decreasing with a non-constant speed, and from September 21, this percentage is zero for pollinators P121, P45, that is to say that the entire amount of "Bisser" is moving towards the "Tamar" stage, however P7 pollinators, P8, P30, P76, P96, P176, Pinvitro and Psami each donated a percentage of "Bisser" respectively equal to 12%, 9%, 20%, 11%, 29%, 26%, 30% and 12.1% by the same date. For the second data palm (Figure 7), the amount of "Bisser" contained in the groups pollinated by P121 and P45 reaches its maximum about 28 August with percentages of "Bisser" equal to 90.83% and 91.26%, respectively, which cancel to September 21. Cook (1959)[27] in a study conducted in California, has demonstrated that temperatures from June 30 to 15 September dates affect the maturation period.

Figure 8 shows that the rate of evolution of fruit to the stadium "Bisser" varies depending on the pollinator and the region, Indeed the percentage of "Bisser" from the P45 is equal to 83.81%, to August 17, for against Pinvitro gave a percentage of "Bisser" equal to 23.6% for the same date.



**Figure 7** Evolution of the Percentage of the Fruit at the Stage "Bisser" According to Pollinators in the Region Degueche 'Palm Tree 2'

At Tamerza site (Figure 8), pollinated fruits P 45, P 121, P 30 and P96 have completed their stage to October 8. While

pollinated fruits with P invitro, P sami, P176, P76, P7 and P8 each gave about the same date a percentage of "bissir" equal respectively to 30.14%, 28.21%, 27.18%, 30%, 20% and 13%. Based on these results we can see that the effect of pollinators on the fruit ripening period is expressed in both sites, some pollinators always seem to be inducers on early stage "Bisser" as P121 and P45 while others are inducers such as tardiness of maturation like P invitro. The effects of other pollinators (P7, P8, P30, P76, P96, P176 and Psami) on fruit ripening are influenced by the regions (climate, soil type.).

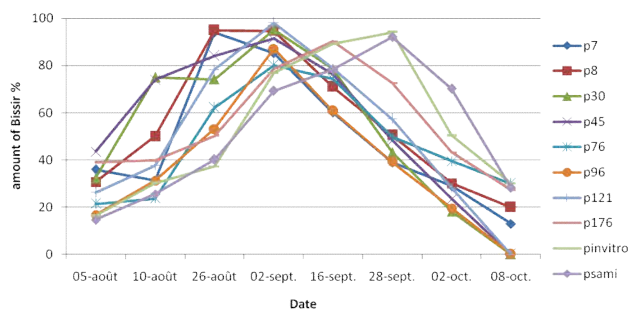


Figure 8 Evolution of the Percentage of the Fruit at the Stage "Bisser" According to Pollinators in the Region Tamerza

**Evolution of Fruit to "Tamar" Step**

early maturation stadium 'Tamar' allows one hand to get a quick availability of dates on national and international markets and also to escape the early autumn rains, this stage begins, depending on the sites and pollinators, 130 days after pollination and lasts for 4-6 weeks to reach full maturity, two weeks more compared to the stadium "Bisser". Unlike two weeks relative to the stage "Bisser" may be the result of climate disruption fall.

According to Figure 9 it can be seen that: At Degueche site for the first palm tree only pollinated fruit with P121, P45, P7 and P8 have completed the stage of maturation to 20 September. While fruits pollinated by P invitro gave about the same date a percentage of "Tamar" equal to 70.1% which is the lowest percentage of "Tamar" while other pollinators provide larger percentages.

As for the 2nd tree (figure 10), only palm fruits pollinated by P45 and P121 have completed the stage of maturation to September 21 and again the fruits pollinated by Pinvitro fail to complete the maturation the same date and has the lowest percentage of "tamar" which is equal to 76.73%.

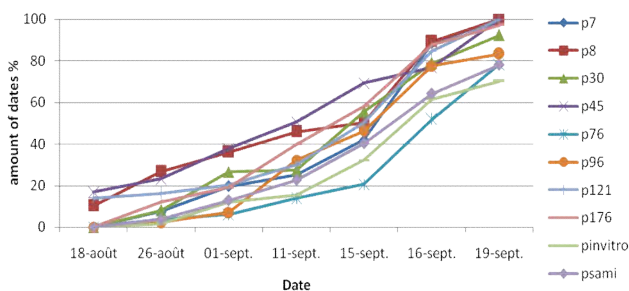


Figure 9 Evolution of the Percentage of the Fruit at the Stage "Tamar" According to Pollinators in the Region Degueche 'Palm Tree 1'

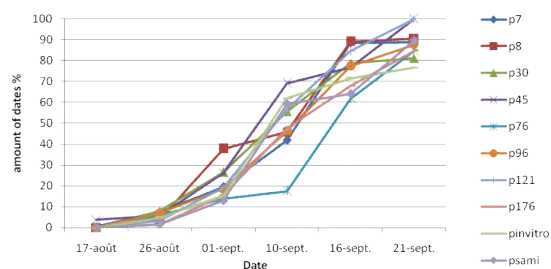


Figure 10 Evolution of the Percentage of the Fruit at the Stage "Tamar" According to Pollinators in the Region Degueche 'Palm Tree 2'

Similarly for the Tamerza region (figure 11) where, the fruits pollinated by P121 and P45 have completed the maturation stage (100 %) to October 8. However, fruit from P invitro pollinators are unable to complete the maturation stage. Indeed, this pollinator at this date (8 October) gives a percentage of 72.82%. The P121 and P45 pollinators have kept the first positions at both stages "Bisser" and "Tamar" in both regions. By against P invitro is an inducer of the lateness of maturity since the maturity dates from these pollinators is not over. For other pollinators, they showed instability in their effects on the maturation dates, hastening or delaying maturity depending on the site that is the case of P30 which has a precocious effect Tamerza and a retarding effect for both tree at Degech site.

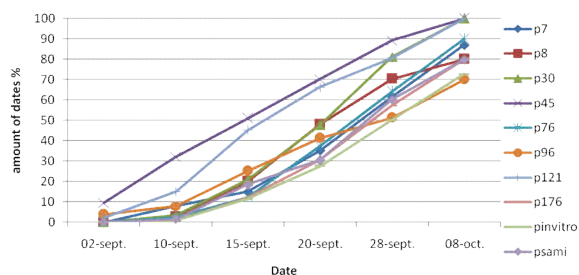


Figure 11 Evolution of the Percentage of the Fruit at the Stage "Tamar" According to Pollinators in the Region of Tamerza

The advance stage "Bisir" leads an advance stage "Tamar". evolution gear changes fruit during stage "Bisser" induced some pollinators have failed to gain a place at the stadium "Tamar". this implies that the fruit passage ds stage "Bissir" stage "Tamar" follows a continuous evolution and changes in climatic conditions affect all fruits, regardless of pollinators [21]. Indeed pollinators that induced early entry dates to the "Bissir" stage are those who have held the top spot at the "Tamar" step. According to Lakhoua (1966) [7], it is the secretion of abscisic acid which is usually induced by a water stress [28]. The variability of response pollinators according to the site depends on the time of installation of the high temperatures and atmospheric vapor pressure deficit resulting from exposure of fruit to the incident radiation [29, 30].

**Study of the Effect of Pollen on Fruit Yield**

**Effect of Pollen on the Caliber of Dates**

The results of the figure 12 show that the fruit size is influenced by the type pollen and site. The average fruit size varies from 3.6 to 4.57 cm long and 1.37 to 1.85 cm wide. The largest fruit in the regions of Degueche and Tamerza are from pollinator Pinvitro inducer of the lateness of the maturation with an

average of 4.40 cm of length and 1.73 cm for wide, followed by the pollinator P121 inducer of earliness of maturity with an average of 4.19 cm for the length and 1.67 cm for wide.

While the smaller fruits are from pollinator 45 inducer of precocity of maturity that gave small fruits in the two regions with an average of 3.9 cm and 1.57cm, The smallest class was as obtained with the fruits pollinated by P sami in both regions. In addition, fruits which were produced in Tamerza were biggest than those product in Degueche whatever the pollinator. The fruit length in Tamarza has varied between 4.05 to 4.57 cm, whereas in Degueche it has hardly exceeded 4.40. The reason for the superiority of Tamarza site can be explained by the admissible maturation dates is delayed by two weeks compared to the dates of Degueche. It has been demonstrated by this work that the pollinator inducer of earliness of maturity (P45) have generally a reducing effect on the caliber of the fruit[24].

Indeed, it was found that fruits 'Dglet Nour' that ripen early to first of October are of lesser quality than those that mature during October and November. Or some male genotypes seem not to respond to this finding in our study the P121 pollinator induction both, precocity and improved caliber. A similar result already obtained in the work of Ben Abdallah *et al.*1986 [8] on the pollinator DB (earliness, improved caliber) and Iqbal *et al.* 2011 [26] who reported a significant effect of different pollens on fruit length.

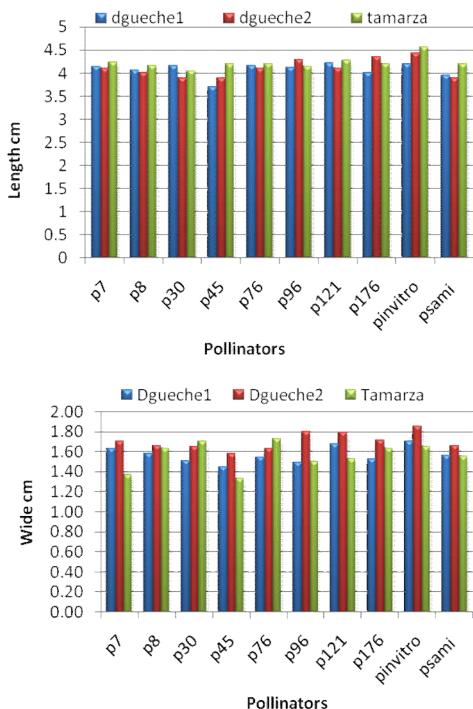


Figure 12 Variation of the length and width of the fruit depending on pollinators and sites

**Effect of Pollen on the Kernel Size**

The results in Figure 13 show that the pollinators inducers of lateness P invitro increase the core size. In the case of early-inducing pollinator (P45) is found a reduction in the caliber of fruit associated with a decrease in core size. Only P121

pollinator that are inducers early detaches these results and provides large fruits as increasing the size of the nuclei.

The effect of pollen on the fruit and kernel size has been shown in the work of Nixon *et al* [31]; Iqbal *et al* 2009 [22] and Shafique *et al.* 2011 [32], found that a direct effect of pollen on fruit and core size.

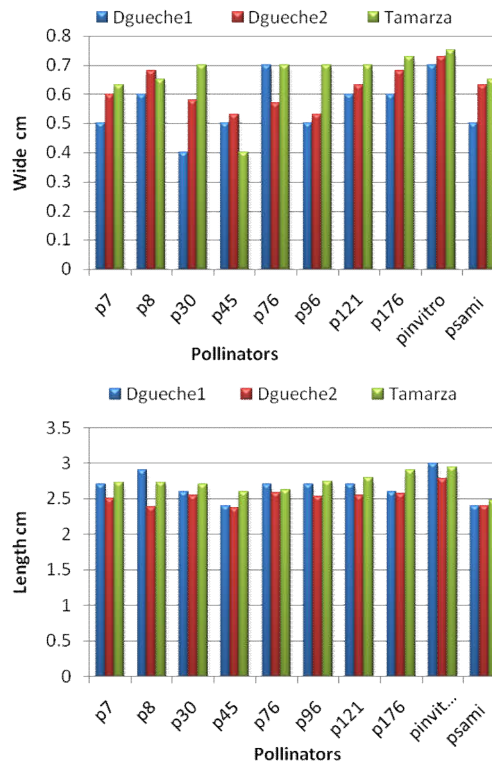


Figure 13 variation of the Length and Width of the Core of the Fruit Depending on Pollinators and Sites

**Effect of Pollen on Fruit Pulp Weight (g)**

Ten fruit of dates is weighed for each type of pollen with 5 repetitions, the results showed that the average weight of fruit varies between 10 and 12 (g), the minimum weight was recorded with the pollinator P 45, however, the weight maximum was recorded with the pollinator P Invitro. The others pollinators showed weight varying between 10 and 11 (g). Figure 14 showed that the weight of fruit also varies depending on the site and it shows that pollination depend also on climate conditions [33].

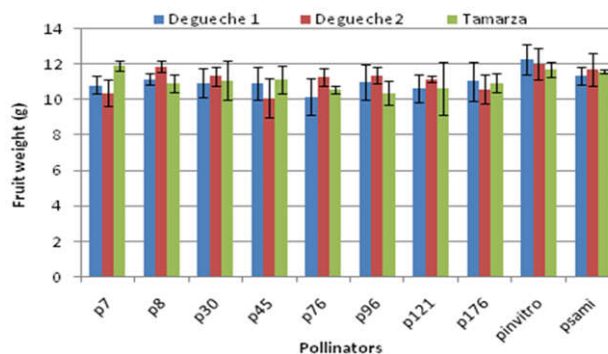


Figure 14 Fruit Weight Variation as a Function of Pollen and Site



The results obtained show that earliness inducers pollinators gave the lowest weight, whereas those that induce maturation lateness showed the largest weight as the P Invitro, however the P121 pollinator is the exception, the P121 pollinator is the exception, although he made induces precocity induces a high fruit weight. it can be concluded that there is an inverse correlation between the time of ripening and fruit weight obtained; more the maturation date is early more the weight of fruit is reduced more the yield is low, while more maturation is delayed more performance fruit is high [34].

## CONCLUSION

In this work, we were able, first, to study the morphology of pollinators 10 based on 51 phenotypic traits. Secondly, we studied the effect of these pollinators on the date of maturity and quality of fruit. The morphological study revealed a significant polymorphism with a similarity index from 0.174 to 0.764 which allows to combine the 10 pollinators into 2 groups. The couple "P176" - "P96" coefficient equal to 0.174 similarity is widely distant morphologically. For against, the highest similarity (0.764) was observed between pollinators "P121" - "P30".

The analysis of the effect of different pollinators of fruit ripening has yielded three groups: one with pollinators hastening fruit ripening: P121 and P45. The second consists of the timer pollinators of fruit ripening: P invitro and the latter consists of pollinators have an unstable effect on ripening. The study of the effect of pollen on to maturity was complemented by a qualitative study from the fruit pollinators studied to compare their qualities. The analysis of the results allowed to classify fruit into three groups: the first with fruits with a high caliber such as P121 and Pinvitro. The second with fruit having a small size as P45 the latter is composed of pollinators that provide fruit having a quality which varies depending on the site such as P7, P30, P176, P76, Psami, P8 and P96.

The identification of a pollinator that induces precocity of fruit ripening with good pomological and biochemical characters of great interest to producers. This work has highlighted the P121 pollinator is the most efficient pollinators among the 10 studied. Indeed, the P121 is ranked in first place as to its ameliorative effect on the quality and the progress of fruit maturation. Moreover, the physiological and molecular phenomenon involved in the differential response of different polen soures also need to be explored. Then establish an identification key to this collection using other pollinators stable and reproducible molecular markers SSR type AFLP

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