

Histological and Physiological Studies on Flowering and Fruiting of Some Peach Cultivars Under Assiut Environment

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THE PRESENT STUDY was carried out during 1997/1998 and 1998/1999 seasons on the peach cvs. (San Pedro, Y9/106 and Rubidoux, as early, mid- and late maturing, respectively grown under Assiut climatic conditions. Histological studies revealed that twelve stages could be detected through the generative process from mid of June up to end Feb or early March. The preliminary stages of floral bud occurred approximately at the same time in the three studies cvs. but the late stages (primordia of ovules) were formed during Jan 26, Feb. 9 and March 9 for the three cvs, respectively.

Chilling units according to total hours $\leq 7.2^{\circ}\text{C}$ were 275, 294 and 323 for San Pedro, Y9/106 and Rubidoux peach cvs, respectively from Dec. till bud swelling at the field. These units were (632, 697 & 837) and (187, 177 & 62) according to total hours $\leq 10^{\circ}\text{C}$ and Utah model, respectively. Concerning heat units for bud break till fruit maturity, it was found that accumulated heat units (GDH) required to budburst, full bloom, fruit set and fruit maturity were higher in Rubidoux cv. than in both San Pedro and Y9/106 cvs.

It could be recommended that more care should be given to peach tree during floral bud differentiation, normally upon harvesting; such care would affect the yield of next year and using heat units calculation as an index for peach fruit maturity. Late cultivar (Rubidoux) is not recommended under Assiut climatic conditions.

Flower bud differentiation is quite important for peach production and it is well known that flowering is an evident important stage in plant life, because of yield

is directly dependent upon its success or failure (Abbas, 1995). The bud formation of peach takes a three period growth, occurring during the fall and early winter, late winter and during the month preceding bloom (Guirgis , 1968, Bazant & Svobada ,1983, Harhash, 1994 and Abbas, 1995). Initiation ended with ovule primodium and for further growth a chilling stimulus during dormancy was required (Lomakin *et al.*, 1974).

Dormancy divided into three type: ecodormancy, paradormancy and endodormancy (Lang *et al.*, 1985). At endodormancy phase, exposure of buds or organs to low temperature less than 7.2°C is required to overcome this type of dormancy (Scalabrelli & Couvillon, 1986, Stino *et al.*, 1986, Smith *et al.*, 1992 and Frisby & Seeley, 1993). Temperatures between 4 and 8°C were effective chilling temperatures, but were less effective than 8°C and temperatures below 4°C were moderately effective (Couvillon ,1995).

The amount of chilling requirement for breaking bud endormancy is differed not only among fruit species, but also between cultivars within the species (usually less than 7.2°C). Additional chilling accumulation will continuously shorten time for budbreak and increase growth degree (Hauagge & Cummins , 1991).

There are many methods used to calculate the amount of chilling and predict the endodormancy completion, (Weinberger, 1967, Richardson *et al.*, 1974, Shaltout & Unrath,1983 and Mansour & Stino, 1986). Dormancy induction and release are regulated by a balance between growth inhibitors and promoters (Powell, 1987, Dennis , 1994 and Lang, 1994).

After completion of endodormancy, the buds will need a heat to develop into shoots and flowers. Several methods can be used to calculate heat requirements and it is calculated from endodormancy completion till various stages of bud development using specified base temperature (Munoz *et al.*, 1986 and Sparks, 1993). Most of these methods accounted it as growing degree hours (GDH) from each hour. The predictive capacity of any method depends on an accurate determination of base temperature. Richardson *et al.* (1974) used 4.4°C as a base temperature of apple and peach buds.

However, before starting any programme for improving peach cvs., accurate
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studies on flowering behaviour must be conducted. Hence, the aim of this study was to study the stages of flower bud differentiation and examine utilization of various models for calculate number of chill units (CU) needed to terminate the endodormancy as well as account number of growing degree hours (GDH) which are required to develop both bud and fruit growth of San Pedro, Y9/106 and Rubidoux peach cultivars (early, mid season and late cvs.) grown under Assiut environments.

Material and Methods

The present investigation has been conducted at the experimental orchard of the Faculty of Agriculture, Assiut University and histological study was proceeded at Botany Dept. The experiment has been extended for two successive seasons 1997/1998 and 1998/1999 on mature bearing trees of three peach cultivars. These cultivars were: San Pedro (Early maturing cv.), Y9/106 (mid-season maturing cv.) and Rubidoux (late maturing cv.).

Five trees-11 years old and spaced 3 m in a square system for each cultivar were randomly selected. The selected trees were approximately vigorously uniform in their vegetative growth. Trees were budded on "Nemagurd" rootstocks and trained as modified leader form. Regular horticultural practices were applied to all experimental trees.

Histological samples

Sampling period: Five buds were taken at random from the fourth node from the base of the current shoots for each cv. The buds were taken at weekly intervals beginning on the first of June, till early February or March (at pink bud stage).

Buds were excised and fixed in F.A.A. solution (Formalin, acetic acid, alcohol 5:5:90, respectively). The buds were transferred from F.A.A. and were dehydrated in a graded series of ethyl alcohol and cleared in xylol. Then, the buds were embedded in paraffin wax at 60°C for three days. Series of sections of 7-10 μm in thickness were prepared using rotary microtome (Leitz 1512). Prior to staining, the mounted sections were deparaffinized in three changes of xylol for 15 min. The sections were later double stained with safranin and light green and mounted by DPX (Sass, 1951). The sections were microscopically examined and photographed on Kodak-colour film.

Field chilling (Meteorological data)

Ten shoots of one-year-old were selected from each tree and tagged according to their similarity in growth to study bud development stages and corresponded GDH.

Orchard temperature every hour for each day from Nov. through August were obtained from the weather station, located at about 100 m from the peach orchard.

Accumulated chilling hours

Both $\leq 7.2^{\circ}\text{C}$ (Chandler *et al.*, 1937) and $\leq 10^{\circ}\text{C}$ (Gilreath & Buchanan, 1979) were used as 1 hr below 7.2 and/or 10°C is equal to one chill unit.

Chilling units- were calculated according to Utah model (Richardson *et al.*, 1974) (Table 1).

TABLE 1. Conversion of hourly temperatures to chilling units according to Utah model.

Utah model	
Temp. ($^{\circ}\text{C}$)	Chill unit contribution
<1.4	0.0
1.5-2.4	0.5
2.5-9.2	1.0
9.2-12.4	0.5
12.5-15.9	0.0
16-18	-0.5
>18	-1.0

Heat units (Growing Degree Hours); GDH

GDH were calculated from the considered date of bud swelling to the various stages of fruit development. The model (using a base temperature of 4.4°C [40°F]) was subtracted from each reading without imposing any limit for higher temperature used (Shallenberger *et al.*, 1959). The accumulated G.D.H. were calculated according to the following equation:

$$\text{G.D.H.} = \sum_{n=1}^{12} (\text{Tm} - 4.4^{\circ}\text{C})$$

where tm = daily temperature per hour.

4.4°C = base temperature

ABA and IAA were determined in peach bud extraction at the central laboratory, Cairo University using Gas Liquid Chromatography (GLC). Extraction was made according to Vogel (1975) and methylation according to the same author. Data are expressed as $\mu\text{g}/1$ g fresh weight.

Results and Discussion

Stages of flower bud differentiation

The behaviour of flower bud differentiation of 3 peach cultivars *i.e.* San Pedro, Y9/106 and Rubidoux as early, midseason and late cultivars, respectively was studied under Assiut climatic conditions. Data presented in Fig. 1-4 describe stages of floral bud differentiation of such cultivars and time of their occurrence.

Twelve stage were found to be needed for peach bud to differentiate. These stages were as follows:

- 1- Vegetative bud formation occurred at the same time, June 19 for the 3 cvs. (Fig. 1A).
- 2- Floral bud flattening occurred on 26/6 in San Pedro and Rubidoux and 2 weeks later in Y9/106 selection (Fig. 1B)
- 3- Formation of sepals primordia on July 9, 3 and 26 for San Pedro (early), Y9/106 (middle) and Rubidoux (late), respectively (Fig. 1C).
- 4- Petal primordium and elongation of sepals occurred on August 11, 26 and Sept. 7 for Sand Pedro, Y9/106 and Rubidoux cvs., respectively (Fig. 1D).
- 5- Formation of Stamens primordia was then proceeded on September 25 (in San Pedro and Rubidoux) but occurred earlier (Sept. 10) in Y9/106 cv., respectively (Fig. 2A).
- 6- Development of the Anther, Filament and the Initiation of Carpel occurred on Oct. 25 in San Pedro, Nov. 8 in Y9/106 and on Dec. 1 in Rubidoux cvs, respectively (Fig. 2B).

- 7- Gynoecium development in Nov. 9, Dec. 1, 12 in the 3 cvs, respectively (Fig. 2C).
- 8- Ovarian cavity appeared in this stage at day 179; from differentiation start of June 19 (Dec. 15), day 192 (Dec. 28) and day 254 (Dec. 28) for the 3 cvs, respectively. Such stage exhibited long period and might cause the delay in the late cv; Rubidoux (Fig. 2D).
- 9- Anther formation with the 2 lobes, then the tetrads formation and finally the formation of pollen grains. Such stage was found to be ended by Feb. 9 in San Pedro, Jan. 19 in Y9/106 and March 9 in Rubidoux cvs. (Fig. 3A).
- 10- Ovules primordia formed during Jan. 26, Feb. 9 and March 9 for the 3 cvs, respectively (Fig. 3B).
- 11- Formation of the outer develop of ovule (integument) appeared on Feb. 9 (day 235), on March 16 (day 270) and on March 23 (day 283) for the 3 cvs. (Fig. 3C), respectively.
- 12- Formation of the inner envelop of ovule (nucellus) started to appear on Feb. 16, March 20 and March 23 for the 3 cvs. (Fig. 3D), respectively.

As a conclusion, the present study exhibited differences among cultivars in the completion of each stage. Difference in carpel primordium formation was a key factor in the delay of late cultivar (Rubidoux); 63 days compared to other cvs *i.e.* differentiation in the late cv. stopped or became minimum for 63 days to start to form carpels. Differences in days of completion the differentiation were 242, 274 and 283 days for the early, middle and late cvs, respectively; *i.e.* middle and late cvs completed their differentiation 32 and 41 days later than the early did; respectively. Such differences may contribute in differences among cvs. budburst which are 12 and 38 days for middle and late cvs. compared to San Pedro cv. (the early one), (Fig. 5).

These results are nearly similar to findings of Guirgis (1968) on Mit-Ghamr and Luttichaw, Harhash (1994) on Redhaven and Fairhaven & Abbas (1995) on Earligrande and Zewayed cvs.

Accumulated chilling

Data illustrated in Table 2 show that the chilling accumulation began from December and throughout early spring. Three models were used to calculate



Fig.1.

Longitudinal sections through the axillary buds of peach cv. San Pedro showing:

- A: The vegetative bud with acute apex
- B: Elongation of the meristematic tissue in the flower bud
- C: Initiation of sepals primordia (Se)
- D: Elongation of the sepals primordia (Se) and beginning of initiation of the petals primordia (Pe).

Longitudinal sections through the axillary buds of peach cv. San Pedro showing:

- A: The vegetative bud with acute apex
- B: Elongation of the meristematic tissue in the flower bud
- C: Swilling of the meristematic tissue and the apex of the flower bud became more flattened. Initiation of the sepal primordium is shown at one side.
- D: Elongation of the sepals primordia (Se) and beginning of initiation of the petals primordia (Pe)

Longitudinal sections through the axillary buds of peach cv. Rubidoux showing:

- A: The vegetative bud with acute apex
- B: Elongation of the meristematic tissue in the flower bud
- C: Initiation of the sepal primordium is shown at one side.
- D: Elongation of the sepals primordia (Se) and beginning of initiation of the petals primordia (Pe)

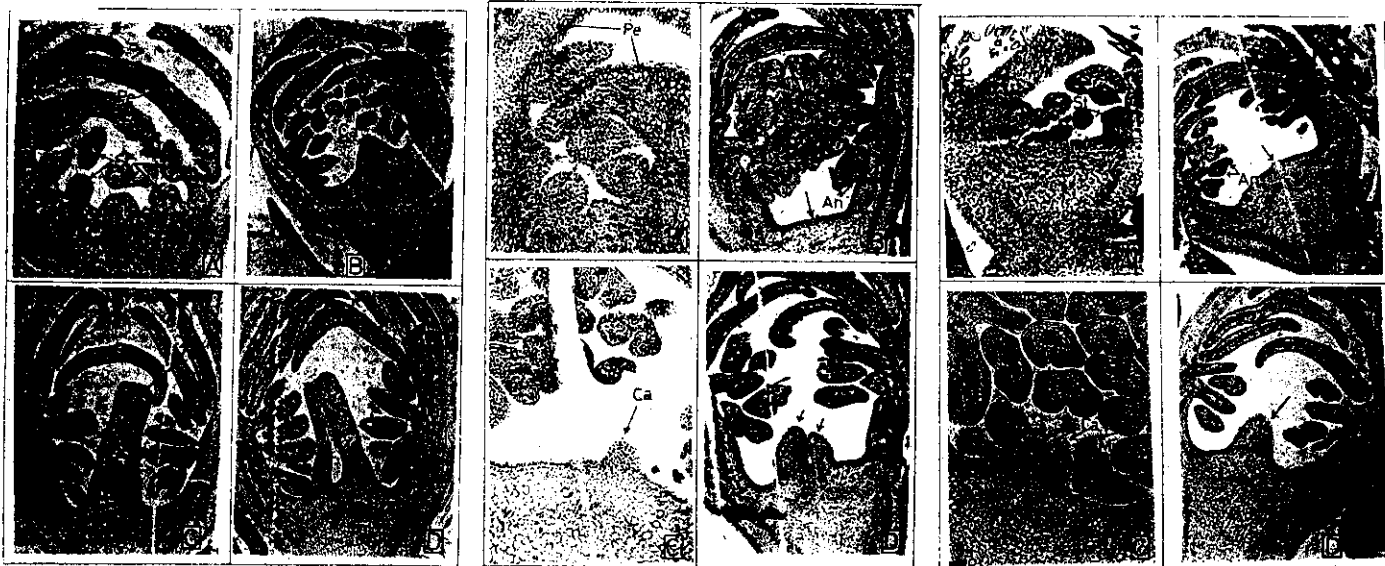


Fig.2.

Longitudinal sections through the axillary buds of peach cv. San Pedro showing:

- A: Elongation of petals primordia (Pe) and initiation of stamen primordium (St).
- B: Development of the anther (An) and filament (F) of the stamen and initiation of the carpel (Ca).
- C: Elongation of carpel.
- D: Appearance of the ovary locale.

Longitudinal sections through the axillary buds of peach cv. Y9/106 showing:

- A: Elongation of petals primordia (Pe) and initiation of stamen primordium (St).
- B: Development of the anther (An) and filament (F) of the stamen. The inside base of the receptacle become more flattened.
- C: Initiation of carpel (Ca) as protuberance on the flattened inside base of the bud.
- D: Elongation of carpel. Appearance of two separated carpels was common in Y9/106 cv.

Longitudinal sections through the axillary buds of peach cv. Rubidoux showing:

- A: Elongation of petals primordia (Pe) and initiation of stamen primordium (St).
- B: Development of the anther (An) and filament (F) of the stamen. The inside base of the receptacle become more flattened.
- C: Initiation of carpel (Ca) as protuberance on the flattened inside base of the bud; and beginning of formation of microsporangia (MI).
- D: Elongation of carpel.

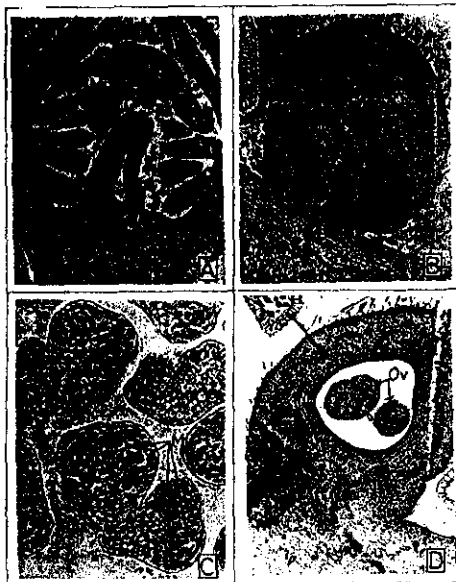
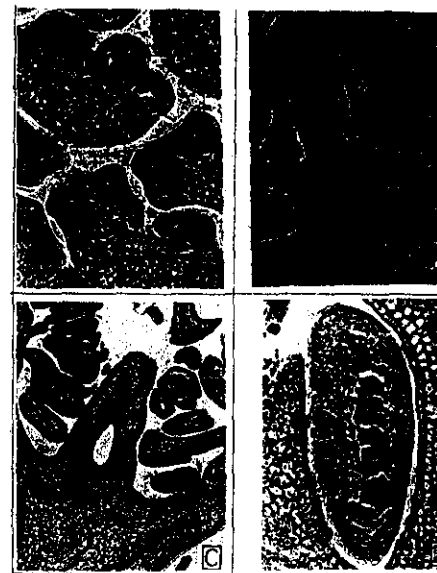


Fig.3.

Longitudinal sections through the axillary buds of peach cv. San Pedro showing:
A:Development of microsporangia (MI)
B:Formation of ovule primordia (Ov).
C:Development of microsporangia (MI); two in each of the two lobes of anther. The sporogenous cells are shown inside the microsporangia.
D:Development of two ovules (OV) inside the carpel locule.



Longitudinal sections through the axillary buds of peach cv. Y9/106 showing:
A:Development of microsporangia (MI); two in each of the two lobes of anther. The sporogenous cells are shown inside the microsporangia.
B: Appearance of the style and stigma, while the pistil base becomes more swelling.
C: Appearance of the ovary locule.
D: Formation of pollen tetrads (Po), they are still enclosed by the wall of spore mother cell.



Longitudinal sections through the axillary buds of peach cv. Rabdoux showing:
A:Development of microsporangia (MI); two in each of the two lobes of anther. The sporogenous cells are shown inside the microsporangia.
B:Appearance of the style and stigma, while the pistil base becomes more swelling.
C: Appearance of the ovary locule.
D: Formation of pollen tetrads (Po), they are still enclosed by the wall of spore mother cell.



Fig.4.

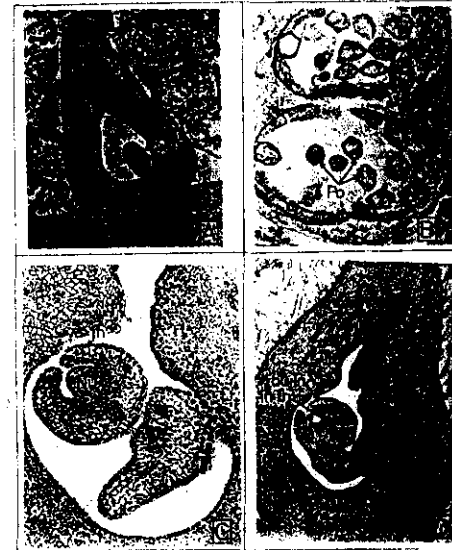
Longitudinal sections through the axillary buds of peach cv. San Pedro showing:

- A: Formation of pollen tetrads (Po).
- B: Mature separated pollen grains (PO).
- C: Beginning the differentiation of the ovule integument (In, one appeared at this time) and development of the nucellus (Nu).
- D: Development of the bitegmic ovule. The outer (Ou, In) and inner (in, In) integuments are shown .



Longitudinal sections through the axillary buds of peach cv. Y9/106 showing:

- A: Formation of ovule primordium (Ov).
- B: Mature separated pollen grains (Po).
- C: Development of two ovules inside the carpel locule. The ovule begin to initiate more close to the pistil base, then by increasing in size the ovules noticed to be attached to the adaxial margin of carpel (compare A with C).
- D: Beginning the differentiation of the ovule integument (In, one appeared at this time) and development of the nucellus (Nu).



Longitudinal sections through the axillary buds of peach cv. Rubidoux showing:

- A: Formation of ovule primordium (Ov).
- B: Mature separated pollen grains (Po).
- C: Development of two ovules inside the carpel locule. The ovule begin to initiate more close to the pistil base, then by increasing in size the ovules noticed to be attached to the adaxial margin of carpel (compare A with C). Beginning the differentiation of the ovule integument (In, one appeared at this time) and development of the nucellus (Nu).
- D: Development of the bitegmic ovule. The outer (Ou, In) and inner (in, In) integuments are showna.

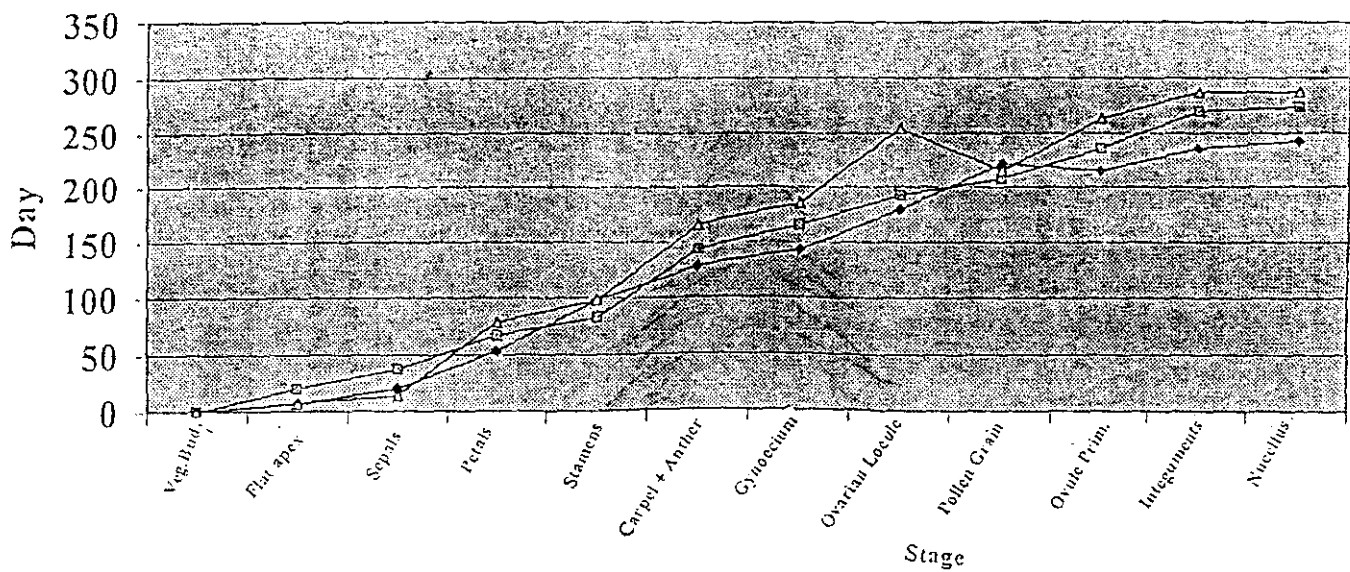


Fig. 5. Flower bud differentiation of San Pedro. Y9/106 and Tubidoux Peach Cultivar.

chilling units from December till bud swelling at the field for San Pedro, Y9/106 and Rubidoux peach cultivars. The first model calculates chilling units according to total hours $\leq 7.2^{\circ}\text{C}$. From such model it can be observed that there were 275, 294 and 323 chilling units from December till bud swelling at the field for San Pedro, Y9/106 and Rubidoux peach cultivars, respectively. The second model calculates chilling units according to total hours $\leq 10^{\circ}\text{C}$. From such model it can be observed that there were 632, 697 and 837 chilling units from December till bud swelling at the field for San Pedro, Y9/106 and Rubidoux peach cultivars, respectively. The third model was Utah model, according to Utah model there were 187, 177 and 62 chilling units for the three cultivars under study. From the obtained data it can be noticed that there was a negation in chilling units according to Utah model. So, such model is not available for Rubidoux peach cultivar, and model 1 & 2 are more suitable for these cultivars.

TABLE 2. Accumulated chill units and date of bud swelling for "San Pedro", "Y9/106" and "Rubidoux" peach cultivars during 1998 and 1999 seasons.

Cultivar	Year	Date of bud swelling	Hours below 7.2°C	Hours below 10°C	Chill units according to Utah model
San Pedro	1997-1998	14/2/1998	264	660	178
	1998-1999	11/2/1999	285	604	194
Mean			275	632	186
Y9/106	1997-1998	25/2/1998	281	742	191
	1998-1999	21/2/1999	307	651	161
Mean			294	697	176
Rubidoux	1997-1998	22/3/1998	324	885	116
	1998-1999	24/3/1999	322	789	7.0
Mean			323	837	62

From this point it could be explained that under warm regions that have insufficient chilling there were series of problems which may collide with some of cultivated deciduous fruit trees. For example, delay and prolongation of the bloom period and foliation with a reduction in the number of flower buds that open, low yields and a delayed harvest.

These previous results are in harmony with those of Mansour & Stino (1986) and Mansour *et al.* (1987) who estimated chill units (C.U.) required to break buds the low chilling requirements peach cultivars, *i.e.* Early Grand and Desert Gold by 223.5 and 255.5 C.U., respectively, under Egyptian conditions according to different models: $\text{hr} \leq 7.2^{\circ}\text{C}$, $\text{hours} \leq 10^{\circ}\text{C}$ and Cairo model.

Heat units (Growing degree hours G.D.H.)

Heat units in terms of growing degree hours (G.D.H.) as correlated with the different stages of flower bud development from the time of rest completion till fruit maturity are explained in Table 3. The accumulated heat units (G.D.H.) as the average of 2 seasons of study; required to reach budburst, full bloom, petal fall, fruit set and fruit maturity for San Pedro, Y9/106 and Rubidoux were 2436, 6409, 8588, 10417, 451.04, 2658, 7979, 10842, 13790, 56653, 3428, 13167, 17189, 21841 and 79804, respectively. Such G.D.Hs for budburst, fruit set and fruit maturity could be fulfilled, under Assiut climatic conditions in 25, 38 and 107 days for San Pedro, 28, 45 and 122 days for Y9/106 and 31, 47 and 132 days for Rubidoux cvs. from the considered time of endodormancy completion, respectively. It could be concluded that there is a positive relationship between chilling requirements and accumulated G.D.H. As previously mentioned by Mansour & Shaltout (1986) and Mansour *et al.* (1987) who stated that GDH were a more accurate method than number of days determining fruit maturity.

TABLE 3. Bud development stage and corresponded GDH during 1998 and 1999 seasons.

Cultivar	Year	Bud burst	Full bloom	Petal fall	Fruit set	Fruit maturity
San Pedro	1998	2438	5884	8182	9552	44436
	1999	2433	6933	8994	11281	45772
Mean		2436	6409	8588	10417	45104
Y9/106	1998	2749	7952	10717	13797	56636
	1999	2666	8005	10966	13783	56670
Mean		2658	7979	10842	13790	56653
Rubidoux	1998	3038	13654	17178	21893	79937
	1999	3818	12680	17200	21789	79670
Mean		3428	13167	17189	21841	79804

Endogenous hormone

Endogenous hormones estimation of peach buds for the San Pedro, Y9/106 and rubidoux cultivars during winter and early spring of 1997/1998 and 1998/1999 seasons is shown in Fig. 1.

Abscic acid content (ABA)

- 1- In general, data indicated higher ABA content in the middle maturing cv. (Y9/106 selection) followed by the other 2 cvs.
- 2- ABA content was found to increase during the dormancy then decreases prior to bud swelling in all cultivars.
- 3- Data of the second season took the similar trend of the first season.

The ABA values of the Y9/106 cv were 4.92, 6.03, 3.5 and 1.4 $\mu\text{g}/\text{g}$ fresh weight during Dec. 1, 1997, Jan. 15, Feb. 15 and March 3, 1998 season. These values were 5.16, 6.3, 3.2 and 2.3 $\mu\text{g}/\text{g}$ f.w. in 1998/1999 season. In San Pedro the early cv, ABA values were relatively lower compared to Y9/106 (middle cv) with the same trend and fluctuation. These values were 0.62, 2.8, 1.2 and 0.8 μg in the first season and 0.94, 2.4, 2.5 and 0.8 μg in the second season. The same trend and fluctuation were found in the late cv. Rubidoux and such values were 1.5, 1.6, 2.1 and 1.22 μg in 1997/1998 season and 2.8, 1.4, 2.04 and 0.88 μg in 1998/1999 season, respectively.

Indol acetic acid content (IAA)

IAA content of Y9/106 cv was found to increase during winter and reached its maximum on Jan. 15 then decreased sharply and reached its minimum by spring. The same trend but with slow decrease was found in the early maturing cv (San Pedro). Different trend was found in the late cv. Rubidoux as IAA decreased from Dec. 1 till Jan. 15 then increased until Spring *i.e.* decrease did not last longer as other cvs.

IAA values were 9.56, 10.22, 1.8 and 0.63 $\mu\text{g}/\text{g}$ fw in Y9/106 1997/1998 season and 3.64, 11.4, 2.1 and 1.8 $\mu\text{g}/\text{g}$ in the second season. In San Pedro cv, these values were 4.2, 5.75, 5.5 and 3.3 μg in the first season and 4.2, 5.3, 5.2 and 2.8 μg in the second one. Meanwhile, such values were 2.75, 2.2, 3.2 and 2.85 μg in the first season of Rubidoux cv. (1.6, 0.71, 2.96 and 3.7 μg in the second season), as shown in Fig. 6.

Such findings would support the early finding of Balandier *et al.* (1993) and Lang (1994) who concluded that dormancy induction and release are regulated by balance between growth inhibitors and promoters.

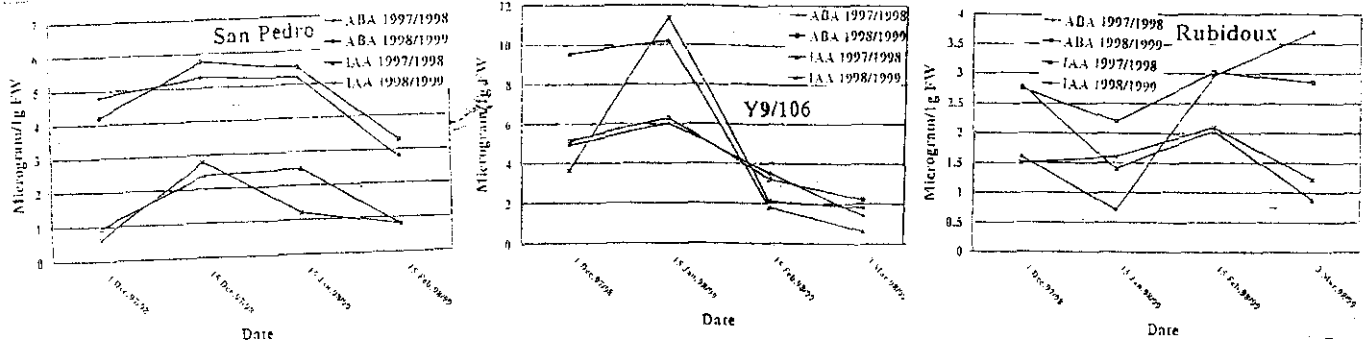


Fig. 6. ABA & IAA in San Pedro, Y 9/106 and Rubidoux peach cultivars.

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دراسات تشريحية وفسولوجية على التزهير والاثمار فى بعض أصناف الخوخ تحت ظروف أسيوط البيئية

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أجريت هذه الدراسة على ثلاثة أصناف من الخوخ هي Sand Pedro (مبكر النضج) ، ١٠٦ ، ٩٠ (متوسط النضج) و Rubidoux (المتأخر النضج) خلال موسمى ١٩٩٧ / ١٩٩٨ و ١٩٩٨ / ١٩٩٩ وذلك بمزرعة كلية الزراعة ومعامل النبات بكلية العلوم جامعة أسيوط . وقد أظهرت الدراسة التشريحية وجود اثنى عشر مرحلة لتكشف البراعم الزهرية للأصناف الثلاثة تبدأ من منتصف يونيه وحتى أواخر فبراير و أوائل مارس .

وقد ظهر أن المراحل الأولى لتكشف البراعم الزهرية حدثت تقريبا فى وقت واحد للأصناف الثلاثة . ولكن المراحل المختلفة تختلف فى موعد حدوثها .

وفيما يتعلق بوحدات البرودة فقد وجد أنها تختلف طبقا للنموذج المستخدم فى حسابها حيث كانت ٢٧٥ و ٢٩٤ و ٣٢٣ وحدة برودة طبقا لعدد الساعات أقل من ٢م^٢م للأصناف San Pedro و ١٠٦ / ٩٠ و Rubidoux على التوالى وذلك من بداية ديسمبر وحتى تفتح البراعم بالحقل . وكانت هذه الوحدات (٦٩٧ ، ٨٢٧) و (١٧٨ ، ١٧٧ ، ٦٢ وحدة) طبقا لعدد الساعات أقل من ١٠م^٢م ونموذج يوتا على التوالى .

وبالنسبة للوحدات الحرارية (G.D.H.) اللازمة للنمو وحتى اكتمال نمو الثمار فقد وجد أنها مرتفعة للصنف Rubidouux عن الصنفين الآخرين .

ومما سبق يوصى بالاهتمام بأشجار الخوخ أثناء تكوين البراعم الزهرية لتحسين محصول الموسم التالي - كذلك استخدام الوحدات الحرارية G.D.H. كمقياس لتحديد اكتمال نمو الثمار وبالتالي موعد الحصاد . كذلك لا يوصى بزراعة الأصناف ذات الاحتياج العالي للبرودة تحت ظروف أسيوط مثل الصنف Rubidouux.