



E-ISSN: 2278-4136  
 P-ISSN: 2349-8234  
 JPP 2018; 7(2): 3721-3728  
 Received: 09-01-2018  
 Accepted: 10-02-2018

**Tsering Dolkar**  
 Division of Fruit Science, Sher-e-Kashmir University of Agricultural Sciences and Technology, Jammu and Kashmir, India

**Mansoor Ali**  
 Division of Soil Science, Sher-e-Kashmir University of Agricultural Sciences and Technology, Jammu and Kashmir, India

**Divya Slathia**  
 Division of Floriculture and Landscape Architecture, Sher-e-Kashmir University of Agricultural Sciences and Technology, Jammu and Kashmir, India

**AB Waheed Wani**  
 Division of Fruit Science, Sher-e-Kashmir University of Agricultural Sciences and Technology, Jammu and Kashmir, India

**Correspondence**  
**Tsering Dolkar**  
 Division of Fruit Science, Sher-e-Kashmir University of Agricultural Sciences and Technology, Jammu and Kashmir, India

## Description of apricot flower and its implications in breeding

**Tsering Dolkar, Mansoor Ali, Divya Slathia and AB Waheed Wani**

### Abstract

Apricot (*Prunus armeniaca* L.) is a species particularly prone to erratic productions and this behaviour has been related to the narrow adaptability of this species. Thus, most apricot cultivars are highly specific in their ecological requirements and low yields are often obtained whenever cultivars are grown in other regions. Flower biology have a close link to fruit set failures in apricot and other fruit trees. Pollen viability and germinability, stigma receptivity, ovule development and longevity, the different factors affecting the effective pollination period (EPP), are reviewed. The study of the inheritance of this and other traits in apricot and other fruit trees has allowed planning of hybridisations to minimise or eliminate the production of undesirable seedlings, increasing the efficiency of the breeding programme. Self-incompatibility is common in apricot cultivars of Central Asian and Irano-Caucasian ecogeographical groups, while cultivars of European group are traditionally considered as self-compatible. The flower biology of apricot have provided valuable information to help select the appropriate parent cultivars for breeding programmes, also this information is transferred to farmers to avoid losses produced by an inadequate cultivar selection.

**Keywords:** effective pollination period, inheritance, male sterility, pistil, pollen, self-(in) compatibility, S-RNases

### Introduction

Apricot (*Prunus armeniaca* L.) is a species particularly prone to erratic productions and this behaviour has been related to the narrow adaptability of this species (Layne *et al.* 1996)<sup>[67]</sup>. Thus, most apricot cultivars are highly specific in their ecological requirements and low yields are often obtained whenever cultivars are grown in other regions. Climatic adaptation is one of the main objectives in most apricot breeding programmes (Hormaza *et al.* 2006), but the causes behind this low adaptability are not clear. The tree is usually around 8-12m tall, with a trunk of approximately 40cm in diameter and a dense, spreading canopy. The slightly tart fruit is very versatile and can be used in a number of culinary ways as well as eaten fresh right off the fruit stand. Fresh apricots are an excellent source of vitamins A, C and E, potassium, and iron, as well as being a great source of beta-carotene. The native range is somewhat uncertain due to its extensive prehistoric cultivation, but is most likely from India, and in Armenia the apricot is considered native as it has been cultivated in the area for many hundreds of years and the species is named *armeniaca*. Today the cultivars have spread to all parts of the globe with climates that support cultivation of the tree and the resulting highly sought-after fruit. Cultivation is generally confined to cool frost-free sites, due to the early blooming but relatively high chilling requirement of the fruit, and fungal disease problems in humid climates. The center of diversity of the apricot is northeastern China near the Russian border (in the Great Wall area). From there it spread west throughout central Asia. Cultivation in China dates back 3000 years. The Romans introduced apricots to Europe in 70-60 BC through Greece and Italy. Apricots probably moved to the US through English settlers on the East Coast, and Spanish Missionaries in California. For much of their history of cultivation, apricots were grown from seedlings, and few improved cultivars existed until the nineteenth century. Cultivars vary among countries, and in Turkey, Iran, Iraq, Afghanistan, Pakistan, and Syria, a great deal of the production is from seedling orchards. Cultivation in the USA was confined to frost-free sites along the Pacific slope of California, due to early bloom but relatively high chilling requirement, and fungal disease problems in humid climates. Now, most of the production in California is in the San Joaquin valley.

### Flower bud density and drop

Depending on the intensity, flower bud drop may negatively influence final yield. Several factors are considered common causes of flower bud drops

(Water stress, lack of chilling, high temperatures during autumn or winter, etc.). Important losses of flower buds have been associated with deficit irrigation treatments in apricot (Uriu, 1964) [110]. However, other authors did not find an influence of different irrigation treatments on flower bud drop in apricot (Albuquerque *et al.* 2003) [3]. Warm temperatures during autumn and winter have been considered responsible for incorrect flower development and, therefore, large flower bud drops in peach (Monet and Bastard, 1971). Unsatisfied chilling requirements have also been related to flower bud drop in apricot (Legave, 1978). However, other authors did not observe an influence of chilling on flower bud drop in apricot cultivars (Albuquerque *et al.* 2003) [3]. The different results found could be explained if flower bud density (number of flower buds per branch section) and flower bud drop were genetically conditioned traits. A strong influence of the cultivar on flower bud drop in apricot has been found (Legave *et al.* 1982) [76]. Also in apricot, when nine different cultivars were studied during three consecutive years, flower bud density and flower bud drop were not affected by the climatic conditions of the different years but there were large differences between cultivars. Flower bud densities ranged from 63 to 180 buds cm<sup>-2</sup> and percentages of flower bud drops were over 50% in many cultivars and ranged from 13% to 72%, expressed as averages from the three years (Albuquerque *et al.*). It has been found also, in peach and nectarine, that flower bud density (Bellini and Gianelli, 1975; Okie and Werner, 1996) [13, 85] is highly dependent on the cultivar studied. A scarce flower bud production and/or high flower bud drop is indicative of poor productivity. Since these characters seem to be cultivar-dependent, they may be inherited and therefore the use of such cultivars as parents within a breeding programme will not be advisable.

### The pollen

Pollen from apricot cultivars it was found that, with the exception of some male sterile cultivars like 'Colorao' or 'Arrogante', most of the apricot cultivars produce pollen in quantities that range from 2,000 to 4,000 grains per anther, which is more than 90,000 pollen grains per flower (Egea and Burgos, 1993) [21]. Furthermore, this pollen has a high percentage of viability and germinates, emitting a pollen tube, in a wide range of temperatures (Vachun, 1981; Egea *et al.* 1992) [111, 39]. Pollen grains placed on the surface of the stigma begin to germinate and elongate in the pollen tubes that grow through the style tissue towards the ovary. The pollen tube wall consists of two main layers of polysaccharide. The inner layer contains predominantly callose or (1.3)- $\beta$ -glucan (Newbigin *et al.* 1993) [84]. Callose layer stained with the fluorochrome aniline blue fluoresces intensely when illuminated with ultraviolet light. The amount of callose is higher in self-incompatible pollen tubes comparing to self-compatible ones. Especially, there is a large deposit of callose close to the swollen tip of the incompatible pollen tubes. Thus, Egea *et al.* 1991 [23] studied that pollen tubes reached the ovary in 48 h, while Guerriero and Bartolini, 1995 [12] reported that, under ideal conditions, they reach the ovary in 48 h, but most often in 72 h. However, according to (Milatović and Nikolić, 2007) [82], 72 h was insufficient for most cultivars, so they extended this period to 120 h. Viti *et al.* 1997 [24] point out that, in apricot, it takes pollen tubes at least 96 h to reach the ovary. Also, Audergon *et al.*, 1999 [9] obtained better results when fixation of pistils was done 96 h rather than 72 h after pollination. In this study, 96 h proved to be enough time to allow compatible pollen tubes to reach the

ovary and ovule. The site of inhibition of pollen tube growth in apricot differs from that normally associated with gametophytic incompatibility. In GSI system, pollen tubes mainly stop their growth in the upper third of the style. However, in our study in most cases we observed that pollen tubes stopped growing in the lower half of the style. Andrés and Durán, 1998 [5] reported that in apricot pollen tubes usually stop their growth in the third quarter of the style length.

### Male sterility

Male sterility is defined as the deviant condition in normally bisexual plants when no viable pollen is formed (Frankel and Galun, 1977) [49]. Male sterility has been exploited as an effective tool to aid hybrid seed production in many crops. However, male sterility is an undesirable characteristic in scion cultivars of *Prunus* to be used for fruit production, because this trait would restrict yield in large monoculture production blocks. Male-sterile cultivars need cross-pollination and production would depend on an adequate pollen transfer from other cultivars. Apricot pollen fertility (Burgos, 1991) [23] in cultivars, 'Arrogante', 'Colorao de Moxó' and 'Colorao' indicated that only three male sterile. Male-sterile anthers can be distinguished visually from normal fertile anthers during the bloom period. Shrunken, discoloured anthers are indicative of male sterility and provide a sharp contrast to the swollen, yellow appearance of normal, pollen-fertile anthers (Burgos and Ledbetter, 1994) [25]. A relatively high number of male-sterile trees were observed from controlled hybridisations among fertile cultivars in apricot and they proposed a preliminary model for the inheritance of the trait. Later, it was confirmed (Burgos and Egea, 2001) [22] that the trait is controlled by one recessive gene (Table 1). Five cultivars or selections included in this study were heterozygous for this trait and, since all hybridisations among them were performed to combine fruit quality attributes and the heterozygous status was unknown, this trait can be of economic importance in the efficiency of an apricot breeding programme, since hybridisation among heterozygous cultivars would produce 25% of male-sterile progeny. These authors found that all crosses between the male-sterile parent and normal cultivars resulted in a completely male-sterile offspring. Furthermore, when these F1 seedlings were open-pollinated or backcrossed with the fertile parent all progenies were male-sterile. The knowledge on the inheritance of this trait will help to plan hybridisations, so that production of male-sterile progeny is avoided through selection of homozygous fertile parents. Also, this information and the progenies generated to obtain it have helped the search for molecular markers for this trait, that will allow detection and elimination of male-sterile plants at the seedling stage (Badenes *et al.* 2000) [11].

### The pistil

It has been demonstrated that fruit set is determined by numerous factors that affect different processes occurring in the pistil during pollination, pollen tubes germination and growth through the stiles and ovule fertilisation. For instance, it has been found that high temperatures during the pre-blossom weeks produce abnormal flowers and diminish fruit set in apricot (Rodrigo and Herrero, 2002) [87] as well as the ovule viability in almond (Egea and Burgos, 1995a) [40]. Stigma receptivity (Egea *et al.*, 1991a; Egea and Burgos, 1992) [23, 39], the role of the pistil in controlling pollen tubes growth (Herrero, 1992; Herrero and Hormaza, 1996) [58, 61],

ovule maturity at anthesis (Egea and Burgos, 1994; Egea and Burgos, 1998; Albuquerque *et al.* 2000 and 2002a) <sup>[25, 43, 1, 2, 24]</sup> and its subsequent evolution (Burgos and Egea, 1993; Burgos *et al.* 1995) <sup>[21, 25]</sup> have been studied widely in apricot and other fruit trees Macro styles. The length of some pistils places the stigmas above the anthers when their natural position should be at the same or a lower height. Macro styles are a cultivar characteristic that is inherited, although climatic conditions, especially temperatures before or after anthesis, play an important role in regulating the manifestation of the trait. In apricot cultivars with the stigma 2 to 3 mm above the anthers, at anthesis, a much lower number of pollen grains has been found on the stigmas than in flowers of cultivars with the anthers at the same height as or above the stigmas, when those flowers were within bagged branches and cross-pollination was absent (Egea and Burgos, 1993) <sup>[21]</sup>. Macro styles may produce important crop failures when there are few bees or when climatic conditions do not allow the activity of these insects. Self-compatible cultivars with long styles may behave as incompatible in these conditions. Also, since stigmas project out of the flower, the risk of quick desiccation and subsequent loss of receptivity is high.

### The stigma

Stigma receptivity is fundamental, in many instances, for explanation of phenomena observed during fruit setting. In some cases, the stigma has been considered responsible for the success of some cultivars like the pear 'Decana del Comizio' (Bini and Bellini, 1971; Bini, 1972) <sup>[14]</sup>. Other papers have reported immature stigmas at anthesis in the pear 'Agua de Aranjuez' (Herrero, 1983; Sanzol *et al.* 2003) <sup>[57, 93]</sup> or the apple 'Cox's Orange Pippin' (Williams *et al.* 1984) <sup>[121]</sup>. In apricot, immature stigmas at anthesis have been found also in some apricot cultivars, reaching an optimum receptivity two to four days after anthesis and losing it very quickly thereafter (Burgos *et al.* 1991; Egea *et al.* 1991) <sup>[23]</sup>. In the Southeast of Spain, many apricot cultivars have an extremely short period in which stigmas are receptive (Egea and Burgos, 1992) <sup>[39]</sup>.

### The ovary and the ovule

The development of the mega gametophyte in relation to fruit set, the occurrence of malformed ovules with degenerated embryo sacs has been observed at different stages of flower development in avocado, olive, apple, pear, cherry, almond, peach and apricot. Frequently, more than two ovules have been found in apricot. However, extra ovules are generally malformed or they degenerate quickly (Burgos and Egea, 1993; Egea and Burgos, 1995) <sup>[21, 40]</sup>. The different embryo sac developmental stages in apricot. At anthesis, apricot ovules are not mature and frequently they are in a very immature stage (Egea and Burgos, 1994; Albuquerque *et al.* 2000 and 2002a) <sup>[25, 1, 2]</sup>. Most ovules examined were within the first three stages of development in our classification (i.e. from ovules without embryo sac to four-nuclei embryo sacs), with high percentages of ovules without a differentiated embryo sac. Lillecrap *et al.* (1999) <sup>[77]</sup> found small and delayed embryo sacs at anthesis in an apricot cultivar with frequent low yields, whereas most embryo sacs had eight nuclei in two other cultivars which produced good yields generally. Apricot cultivars with immature ovules at anthesis (embryo sacs with four nuclei) produced normal crops (Egea and Burgos, 1998) <sup>[43]</sup>. Therefore, those ovules with, at least, a four-nuclei embryo sac at anthesis have been considered as functional (Albuquerque *et al.* 2002a) <sup>[2]</sup>. The percentages of

functional ovules and fructification of nine apricot cultivars are reported. Cultivars with more than 50% fruit set had also high percentages of functional ovules, suggesting that a certain degree of mega gametophyte development at anthesis is necessary for fertilisation to be successful, although it may not be enough to ensure a good crop since some cultivars with high percentages of functional ovules had low fruit set. Both the ovary and the ovule provide signals that orient and direct pollen tube growth to the right course (Herrero, 2001) <sup>[60]</sup>. In peach, particular secretions from ovary cells along the pollen tube pathway are required for the pollen tube to proceed towards the embryo sac (Arbeloa and Herrero, 1987; Herrero, 2000) <sup>[58, 59]</sup>.

### The effective pollination period

Williams (1966) <sup>[117]</sup> introduced the concept of effective pollination period (EPP) as the period during which pollination is effective to produce a fruit, and described in detail the approach used to estimate the EPP in orchard conditions, which basically consists of hand-pollinating flowers at time intervals from anthesis and later recording the initial and final fruit set in these flowers (Williams, 1970a). Microscopic examination of pollen tube kinetics and ovule viability can be useful as an indirect estimation of the EPP. Since the EPP is determined by the longevity of the ovule minus the time required by the pollen tube to reach the ovule, this indirect estimation will be valid whenever the EPP values do not exceed the stigmatic receptivity period (Williams, 1966). The microscopic approach provides additional information on the parameters that limit the EPP that is not obtained with the estimation in the orchard. The EPP was defined as a function of pollen tube speed and ovule longevity. Therefore, it links female fertility and pollination and is an expression of the likelihood that the flowers set fruit. Flower fertility is the capability to produce fruits when flowers are pollinated, at the right time, with compatible pollen. Theoretically, each normally-developed flower is able to set a fruit if pollinated with the appropriate pollen just after anthesis. Its failure to do so is indicative of female sterility. However, under normal conditions, flowers are not always pollinated at anthesis and stigmas remain receptive for several days (Williams, 1970b; Williams *et al.*, 1984). Stigma receptivity, the speed of pollen tube growth and ovule longevity are three factors commonly-studied in the literature about EPP. Different studies report their relative importances, depending on the species and climatic conditions. There must be a good synchronisation between them, although genetic and environmental factors may unbalance the process and, therefore, decrease fruit setting (Thompson and Liu, 1973). In fruit trees, including apricot, EPP duration has been estimated to be very variable, depending on the species, cultivar and environmental conditions, ranging from two days to more than a week (Sanzol and Herrero, 2001) <sup>[92, 60]</sup>. When the limiting factor of EPP was determined, in the reviewed papers, a good correlation was found between the two period lengths. In kiwi, the short EPP found was attributed to a fast loss of pollen germinability due to high temperatures (Galimberti *et al.*, 1987) or to lack of support of pollen germination by the stigma (González *et al.*, 1995). Delays in stigma maturation (Martínez-Tellez and Crossa-Raynaud, 1982; Herrero, 1983) <sup>[57]</sup>, or a short receptivity period (Williams, 1965; Guerrero-Prieto *et al.*, 1985; Burgos *et al.*, 1991; Egea *et al.* 1991a) <sup>[23]</sup> may limit the EPP. Williams (1970c) found that ovule development is affected by high temperatures, but with temperatures between 7 and 15°C

ovule development is normal while there is an increase in the speed of pollen tube growth. In these conditions, the EPP is improved. In the climatic conditions of South-Eastern Spain, the limited period of stigma receptivity has been found to be responsible for a short EPP in apricot. For many cultivars examined, high temperatures at bloom limit the stigma receptivity to only one to three days after anthesis, in the most extreme cases (Egea *et al.* 1991; Egea and Burgos, 1992) [23, 39]. Pollen tubes grow fast in these conditions but at least three days are necessary to reach the ovary. The longevity of the ovule is related to its stage of development at anthesis. Ovules mature at anthesis will remain viable only a short time, limiting the EPP. On the other hand, if ovules are very immature at anthesis, there may be asynchronies between pollen tube arrival and the maturity of the ovules, which will affect fruit set. The most favourable condition for fructification would be when ovules are at intermediate stages of development (embryo sacs with four to eight nuclei) at anthesis (Alburquerque *et al.* 2002a) [2].

**Table 1:** Percentages of functional ovules and fruit set in different apricot cultivar

Cultivar	Functional ovules	Fruit set
Palstein	84.9 ± 4.92	53.8 ± 1.90
Priana	73.7 ± 7.14	56.4 ± 4.55
Beliana	90.9 ± 4.33	63.5 ± 0.40
Colorao	74.3 ± 7.00	7.5 ± 1.67
Guillermo	32.3 ± 8.02	23.5 ± 0.42
Goldrich	84.6 ± 5.78	16.6 ± 6.66
Pepito	22.1 ± 3.01	12.0 ± 3.02
Bergeron	15.2 ± 2.98	12.8 ± 3.25
Modesto	12.5 ± 5.23	11.0 ± 3.40

Source (Alburquerque *et al.* 2002a) [2]

### Self-incompatibility

Incompatibility is the inability of a fertile seeded plant to produce zygotes after self- or cross-pollination (self- or cross-incompatibility) (Heslop-Harrison, 1975). This reaction is an active, regulated constraint of pollen tube growth where, depending on the species and the system operating, the process may be blocked at the initial steps of pollen hydration and germination on the stigma (Dickinson, 1995), during pollen tube growth in the style (Matton *et al.*, 1994) or further down in the ovary (Sage *et al.*, 1994). Recognising and rejecting their own pollen before fertilisation allows self-incompatible plants to promote outcrossing and improve genetic variability, which is considered to play an important role in the evolutionary success of the angiosperms. Outcrossing establishes a regulated degree of heterozygosity in the population. Incompatibility occurs in more than 3,000 species of 250 genera, that belong to about 70 families (Van Gestel, 1976). Although, traditionally, the European group of apricot (within which the apricots grown in Europe, North America, South Africa and Australia are included) has been described as self-compatible (Mehlenbacher *et al.*, 1991), in the last two decades many widely-cultivated apricot cultivars have been described as self-incompatible. In fruit trees, incompatibility complicates horticultural practices because self-incompatible clones require the addition of pollination nators and the yield depends on abundant pollen transfer among the trees.

### Genetic control

In *Prunus*, the incompatibility system operating in most of the studied species is controlled by one gene with several different alleles. Pollen is rejected when its S-allele is present

in the genotype of the style. Hence, an incompatibility reaction will occur between two plants if their genotypes at the S locus do not differ in at least one allele (De Nettancourt, 1972; Heslop-Harrison, 1975). Sweet cherry was the first *Prunus* species where this model was described (Crane and Brown, 1937). The same mechanism has been demonstrated in almond (Dicenta and García, 1993) and apricot (Burgos *et al.* 1997) [24]. However, a different mode of inheritance was found in Japanese plum, for which it has been proposed that two genes with epistatic relationships control the trait (Arora and Singh, 1990). In apricot, alleles for self-compatibility would allow pollen tube growth in any style. Self-incompatibility alleles would stop pollen tube growth if the same allele was present in the pistil and the pollen. Dicenta and García, 1993 determine the mode of inheritance of self-(in)compatibility in apricot, 19 families with a total of 948 seedlings/ Seedling segregation for the trait allowed it to be deduced that the parents used were heterozygous. Also, there were two families where segregation could only be explained if the parents shared one allele. A similar situation had been found previously in almond when crossing 'Ferragnes' with the self-compatible cultivars 'Genco' and 'Tuono' Further work on stelar proteins of almond (Boskovic *et al.*, 1997) and apricot (Burgos *et al.* 1998) [43] cultivars found the existence of a common S-allele. It could only happen if both self-incompatible parents have the same genotype. Two groups of cross-incompatible cultivars have been described after controlled pollinations. One of them includes three Hungarian apricot cultivars (Nyéki and Szabó, 1995) and the other the North American cultivars 'Lambertin', 'Goldrich' and 'Hargrand' (Egea and Burgos, 1996) [42].

### Molecular aspects of incompatibility

Within the Rosaceae, a correlation between known genotypes for self-(in) compatibility and bands resulting from electrophoresis of stelar extracts has been found in Japanese pear (Hiratsuka *et al.*, 2001; Sassa *et al.*, 1992) where the proteins have been characterised as glycoproteins with RNase activity (Hiratsuka, 1992; Sassa *et al.*, 1993; Hiratsuka *et al.*, 1995; Hiratsuka and Okada, 1995). Similar results have been found in apple (Sassa *et al.*, 1994), and European and Chinese pears (Tomimoto *et al.*, 1996). In *Prunus*, similar studies have been carried out in sweet cherry (Mau *et al.*, 1982; Boskovic and Tobutt, 1996; Boskovic and Tobutt, 2001) and almond (Tao *et al.*, 1997; Boskovic *et al.*, 1997; Certal *et al.*, 2002). It was found that these proteins were inherited as if they were the products of the S gene (Burgos *et al.* 1998) [43] and this methodology was used to genotype unknown cultivars and selections from the breeding programme (Alburquerque *et al.* 2002b) [4]. A further step in the molecular research on S-alleles in fruit trees was the use of a combination of S-allele-specific primers, designed from non-conserved sequences from each allele in apple, and the digestion of PCR products with S-allele-specific restriction enzymes (Janssens *et al.*, 1995). The identification of S-alleles correlated perfectly with information on genotypes from phenotypic and RNases analyses and it is a rapid and useful method for determination of the genotype of different apple cultivars (Sakurai *et al.*, 1997 and 2000). A recent paper reports the identification of 15 different S-alleles in apple using this methodology (Broothaerts, 2003). The same strategy, with or without modifications, has been used to design specific primers for S-alleles in almond (Tamura *et al.*, 2000; Channuntapipat *et al.*, 2003), pear (Zuccherelli *et al.*, 2002), sweet cherry (Tao *et al.*, 1999; Yamane *et al.*, 2000; Sonneveld *et al.*, 2001; Wiersma

*et al.*, 2001) and Japanese apricot (Yaegaki *et al.*, 2001). In apricot, the alleles S1 and S2 have been sequenced completely (Romero *et al.*, 2003) by using a bacterial artificial chromosome (BAC) library from the cultivar Goldrich (Vilanova *et al.*, 2003). This is a first step that will allow the design of primers from these sequences in order to amplify different S-alleles in apricot. The possibility of designing primers for the self-compatibility allele found in all self-compatible apricot cultivars tested to date (Albuquerque *et al.* 2002b)<sup>[4]</sup> is especially interesting (Burgos *et al.* 1998)<sup>[43]</sup>. A similar strategy has allowed the design of molecular markers for this important trait in Japanese apricot (Tao *et al.* 2000, 2002a and 2002b).

### Conclusions

The study of the flower biology of apricot, described in this review, has had strong implications for the breeding programme of this species. First of all, the knowledge of the factors limiting fruit set in an important number of commercial cultivars has oriented the selection of parents. Some cultivar-dependent characteristics, like macro styles and flower bud density or drop, indicate that some cultivars would not be a good choice as parents in the breeding programme. Other factors, like ovule immaturity at anthesis, are signs of bad adaptation of the cultivars to local climatic conditions and these, therefore, would also be a wrong parental selection. In those cases when such parents must be used, the knowledge of these characteristics is important in order to evaluate the seedlings, paying much attention to the possible segregation of these traits within the progenies in order to select the ones that have not inherited the undesirable characters. Determining the mode of inheritance of economically-important traits improves the efficiency of breeding. For instance, male sterility may produce up to 25% of male-sterile seedlings from crosses between fertile heterozygous cultivars. The selection of the appropriate parents is, again, the solution. Also, determining the inheritance of self-(in) compatibility and the parents' genotypes for this trait allows hybridizations to be planned which minimize or eliminate the production of self-incompatible seedlings. The correlation between stylar RNases and different S alleles has been a great advance for determination of the genotype of a good number of cultivars. With this methodology, homozygous self-compatible cultivars can be easily identified, which will produce 100% self-compatible progeny regardless of the other parent's genotype. If the necessity of evaluating the progenies generated within the breeding programme, to discard the self-incompatible seedlings, is eliminated, the programme is speeded up, which greatly reduces its cost. Self-incompatibility phenotype determination by controlled crosses and evaluation of fruit set or pollen tube growth as well as RNase analysis, to determine the genotype at the S locus, need mature trees with flowers, which, for fruit trees, means at least three years after seeds are obtained. Using PCR with S-allele-specific primers allows detection of the self-incompatible genotype in the first stages of plant development, and therefore allows roguing of undesirable seedlings straight after germination of the seeds. Specific primers to amplify selectively the allele (or alleles) that determine self-compatibility are molecular markers for this trait with 100% efficiency, since they are located within the S locus. In apricot, these primers have not yet been identified nor efficient molecular markers developed. This interest is, possibly, closely linked to the fact that this knowledge may avoid production failures and also allows the efficiency of the fruit breeding programmes to be increased.

### References

1. Albuquerque N., Burgos L., Egea J. Consequences to fertilization of the developmental stage of apricot ovules at anthesis. *J. Hortic. Sci. Biotech.* 2000; 75: 662-666.
2. Albuquerque N., Burgos L., Egea J. Variability in the developmental stage of apricot ovules at anthesis and its relationship with fruit set. *Ann. Appl. Biol.* 2002; 14: 147-152.
3. Albuquerque N., Burgos L., Egea J. Apricot flower bud development and abscission related to chilling, irrigation and type of shoots. *Sci. Hortic. Amsterdam.* 2003; 98: 265-276.
4. Albuquerque N., Egea J., Pérez-Tornero O., Burgos L. Genotyping apricot cultivars for self (in) compatibility by means of RNases associated with S alleles. *Plant Breeding.* 2002; 121: 343-347.
5. Andrés M.V., Durán J.M. Self-incompatibility in Spanish clones of apricot (*Prunus armeniaca* L.) tree. *Euphytica.* 1998;101:349-355.
6. Arbeloa A., Herrero M. Valoración de la translocación al óvulo y de la esterilidad femenina en melocotonero. *Anales de la Estacion Experimental de Aula Dei.* 1985; 17: 214-220.
7. Arora R.L., Singh R. Genetics of incompatibility in plum (*Prunus salicina* Lindl). *Ind. J. Hort.* 1990; 47: 1-11.
8. Audergon J.M., Signoret V., Duffillol J.M., Gilles F. Les variétés d'abricot. *L'Arboriculture Fruitière.* 1988; 403: 22-56.
9. Audergon J.M., Guerriero R., Monteleone P., Viti R. Contribution to the study of inheritance of the character self-incompatibility in apricot. *Acta Hortic.* 1999; 488:275-280.
10. Austin P.T., Hewett E.W., Noiton D., Plummer J.A. Self incompatibility and temperature affect pollen tube growth in 'Sundrop' apricot (*Prunus armeniaca* L.). *J. Hortic. Sci. Biotech.* 1998;73: 375-386.
11. Badenes M.L., Hurtado M.A., Sanz F., Archelos D.M., Burgos L., Egea J., Llácer G. Searching for molecular markers linked to male sterility and self-compatibility in apricot. *Plant Breeding.* 2000; 119:157-160.
12. Bassi D., Guerriero R., Pennone F. Le cultivar di albicocco. *Riv. Frutt.* 1995; 2: 39-42.
13. Bellini E., Gianelli G. Sul valore tassonomico di alcuni caratteri del ramo nel pesco. *Riv. Ortoflorofrutt It.*1975; 59: 440-458.
14. Bini G., Bellini E. Macrosporogenesi nella cultivar di pero 'Decana del Comizio' e ricerca del periodo più opportuno per la sua impollinazione. *Riv. Ortoflorofrutt It.* 1971; 55(4): 332-336.
15. Blane A., Broquaire J.M., Chanffour D., Clauzel G., Duffillol J.M., Giard A., Gilles F., Moulon B., Audergon J.M., 2004. Soledane, Florilege, Bergarouge, three new apricot cultivars for French country. *Acta Hort.* (In Press).
16. Boskovic R., Tobutt K.R. Correlation of stylar ribonuclease zymograms with incompatibility alleles in sweet cherry. *Euphytica.* 1996; 90: 245-250.
17. Boskovic R., Tobutt K.R. Genotyping cherry cultivars assigned to incompatibility groups, by analysing stylar ribonucleases. *Theor. Appl. Genet.* 2001;103: 475-485
18. Boskovic R., Tobutt K.R., Batlle I., Duval H. Correlation of ribonuclease zymograms and incompatibility genotypes in almond. *Euphytica.* 1997; 97: 167-176.

19. Broothaerts W. New findings in apple S-genotype analysis resolve previous confusion and request the re-numbering of some S-alleles. *Theor. Appl. Genet.* 2003; 106:703-714.
20. Brown D.S. The relation of temperature to the flower bud drop of peaches. *Proc. Amer. Soc. Hortic. Sci.* 1958;71: 77-87.
21. Burgos, L. Biología floral de variedades de albaricoquero (*Prunus armeniaca* L.). Doctoral Thesis. Universidad de Murcia. BURGOS L., BERENGUER T., EGEE J., 1993. Self- and cross-compatibility among apricot cultivars. *Hort. Science.* 1991;28:148-150.
22. Burgos L., Egea J., 2001. Inheritance of male sterility in apricot. *Int. J. Hort. Sci.* 7, 12-14.
23. Burgos L., Egea J., Dicenta F. Effective pollination period in apricot (*Prunus armeniaca*, L.) varieties. *Ann. Appl. Biol.* 1991; 119: 533-539.
24. Burgos L., Egea J., Guerriero R., Viti R., Monteleone P., Audergon J.M. The self-compatibility trait of the main apricot cultivars and new selections from breeding programmes. *J. Hortic. Sci.* 1997; 72: 147-154.
25. Burgos L., Ledbetter C.A. Observations on inheritance of male sterility in apricot. *Hort. Science.* 1994; 29:127.
26. Burgos L., Ledbetter C.A., Pérez-Tornero O., Ortín-Párraga F., Egea J., 1997. Inheritance of sexual incompatibility in apricot. *Plant Breeding.* 116, 383-386.
27. Burgos L., Pérez-Tornero O., Ballester J., Olmos E. Detection and inheritance of stilar ribonucleases associated with incompatibility alleles in apricot. *Sex Plant Reprod.* 1998;11:153-158.
28. Ceta. L'ouest Américain. L'abricotier. L'Arboriculture Fruitière. 1990;426:55-58.
29. Cappellini P., Limongelli F. Indagine sull'autofertilità delle piu importanti cultivar di albicocco vesuviane. *Ann Ist Sper Fruttic XII.* 1981; 47-55.
30. Certal A.C., Almeida R.B., Boskovic R., Oliveira M.M., Feijó J.A. Structural and molecular analysis of self-incompatibility in almond (*Prunus dulcis*). *Sex Plant Reprod.* 2002;15: 13-20.
31. Channuntapipat C., Wirthensohn M., Ramesh S.A., Batlle I., Arús P., Sedgley M., Collins G.G. Identification of incompatibility genotypes in almond (*Prunus dulcis* Mill.) using specific primers based on the introns of the S-alleles. *Plant Breeding.* 2003;122: 164-168.
32. Crane M.B., Brown A.G. Incompatibility and sterility in the sweet cherry (*Prunus avium*, L). *J. Pomol.* 1937;15: 86-116.
33. Crossa-Raynaud P. Situation de la production de l'abricotier en Europe et Afrique du Nord. In: Journées Nationales de l'abricotier at Perpignan. Ed Menard, Toulouse, pp. 1961;53-54.
34. De Nettancourt D. Self-incompatibility in basic and applied researches with higher plants. *Gen. Agr.* 1972; 26:163-216.
35. Dicenta F., García J.E. Inheritance of self-compatibility in almond. *Heredity.* 1993;70, 313-317.
36. Dickinson H.G. Dry stigmas, water and self-incompatibility in Brassica. *Sex Plant Reprod.* 1995;8:1-10.
37. Egea J., Garcia J.E., Egea L., Berenguer T. Self-incompatibility in apricot cultivars. *Acta Hort.* 1991;293: 285-293.
38. Egea J., Burgos L. Effective pollination period as related to stigma receptivity in apricot. *Sci. Hortic-Amsterdam.* 1992;52:77-83.
39. Egea J., Burgos L. Producción de polen y autopolinización en albaricoquero (*Prunus armeniaca* L.). *Proc. of II Rencontres sur l'abricotier Avignon* (France), 27-31 May 1991. (Audergon J.M., ed). Ed Commission des Communautés Européennes. Montfavet, 1993; 21-29.
40. Egea J., Burgos L. Double keneled fruits in almond (*Prunus dulcis*, Mill.) as related to pre-blossom temperatures. *Annual of Applied Biology* 1995; 126:163-168.
41. Egea J., Burgos L. Supernumerary ovules in flowers of apricot. *Acta Hort.* 1995b;384: 373-377.
42. Egea J., Burgos L. Detecting cross-incompatibility of three North American apricot cultivars and establishing the first incompatibility group in apricot. *Journal of American Society Horticulture Science* 1996; 121:1002-1005.
43. Egea J., BURGOS L. Fructification problems in continental apricot cultivars growing under Mediterranean climate. Ovule development at anthesis in two climatic areas. *J. Hortic. Sci. Biotech.* 1998; 73:107-110.
44. Egea J., Burgos L., García J.E., Egea L. Stigma receptivity and style performance in several apricot cultivars. *Journal of Horticulture Science* 1991a; 66:19-25.
45. Egea J., Burgos L., Zoroa N., Egea L. Influence of temperature on the in vitro germination of pollen of apricot (*Prunus armeniaca* L.). *J. Hortic. Sci.* 1992; 67(2):247-250.
46. Egea J., Dicenta F., Burgos L. 'Rojo Pasión' apricot. *HortScience* (In Press). 238 L. Burgos *et al.* / *Span. J. Agric. Res.* 2004a; 2 (2): 227-241.
47. Egea J., Dicenta F., Martínez-Gómez P., Burgos L. 'Selene' apricot. *HortScience* (In Press). 2004b.
48. Egea J., García J.E., Egea L., Berenguer T. Self-incompatibility in apricot cultivars. *Acta Hort.* 1991b; 293: 285-293.
49. Frankel R and Galun E. Pollination mechanisms, reproduction and plant breeding. In: *Monographs on theoretical and applied genetics*, 2. Ed Springer-Verlag, Berlin-Heidelberg-New York, 1977.
50. Galimberti P., Marro M., Youssef J. Periodo utile d'impollinazione in *Actinidia chinensis* (Planch). *Riv Frutt.* 1987; 1:51-54.
51. García J.E., Egea J., Egea L., Berenguer T. The floral biology of certain apricot cultivars in Murcia. *Adv Hort Sci.* 1988; 2: 84-87.
52. Glucina P.G., Bristol R.W., Lewis I.K. Cluthagold: A promising new late season apricot. *Orchardist of New Zealand.* 1988;61:37.
53. Glucina P.G., Hosking G., Mills R. Evaluation of apricot cultivars for Hawke's Bay. *The Orchardist.* 1990;2:21-25.
54. Guerrero-Prieto V.M., Vasilakakis M.D., Lombard P.B. Factors controlling fruit set of 'Napoleon' sweet cherry in western Oregon. *Hort. Science.* 1985; 20: 913-914.
55. Guerriero R. and Bartolini S. Flower biology in apricot: main aspects and problems. *Acta Hort.* 1995;384:261-271.
56. Gulcan R., Askin A. A research on the reasons of unfruitfulness of *Prunus armeniaca* cv. Tokaloglu. *Acta Hort.* 1991; 293: 253-257.
57. Herrero M. Factors affecting fruit set in 'Agua de Aranjuez' pear. *Acta Hort.* 1983;139: 91-96.
58. Herrero M. From pollination to fertilization in fruit trees. *Plant Growth Regul.* 1992; 11: 27-32.

59. Herrero M. Changes in the ovary related to pollen tube guidance. *Ann Bot.* 2000; 85: 79-85.
60. Herrero M. Ovary signals for directional pollen tube growth. *Sex Plant Reprod.* 2001; 14: 3-7.
61. Herrero M, Hormaza JI. Pistil strategies controlling pollen tube growth. *Sex Plant Reprod.* 1996; 9: 343-347.
62. Heslop-Harrison J. Incompatibility and the pollen-stigma interaction. *Ann Rev Plant Physiol.* 1975; 26: 403-425.
63. Hiratsuka S. Characterization of an S-allele associated protein in Japanese pear. *Euphytica.* 1992; 62: 103-110.
64. Hiratsuka S., Okada Y. Some properties of a stylar protein associated with self-incompatibility genotype of Japanese pear. *Acta Hort.* 1995; 392: 257-264.
65. Hiratsuka S, Zhang SL, Nakagawa E, Kawai Y. Selective inhibition of the growth of incompatible pollen tubes by S- protein in the Japanese pear. *Sex Plant Reprod.* 2001; 13: 209-215.
66. Janssens G.A., Goderis I.J., Broekaert W.F., Broothaerts W. A molecular method for S-allele identification in apple based on allele-specific PCR. *Theor Appl Genet.* 1995; 91:691-698.
67. Layne REC, Bailey CH, Hough LF: Apricots. In: Janick, J, Moore, JN., (eds.), *Fruit breedings Volume I: Tree and Tropical Fruits*, 79-109. John Wiley & Sons Inc. New York, Chichester, Brisbane, Toronto, Singapore, 1996.
68. Hormaza J.I., Yamane H., Rodrigo J: Apricot. In: Kole, C. (ed.), *Genome Mapping & Molecular Breeding.* 2007; 7:173-189.
69. Lamb R.C., Stiles W.C. Apricots for New York state. *New York Food and Life Sciences Bulletin.* 1983; 100:1-4.
70. Ledbetter CA. Notice to fruit growers and nurserymen relative to the naming and release of the Helena apricot cultivar. *Cultivar release notice, A.R.S. (U.S.D.A.).* 1994.
71. Ledbetter C.A. Notice to fruit growers and nurserypersons of the release of Lorna apricot. *Cultivar release notice, A.R.S. (U.S.D.A.).* 1998.
72. Ledbetter C.A. Notice to fruit growers and nurserypersons of the release of Apache apricot. *Cultivar release notice, A.R.S. (U.S.D.A.).* 2002.
73. Ledbetter C.A. Notice to fruit growers and nurserypersons of the release of Nicole apricot. *Cultivar release notice, A.R.S. (U.S.D.A.).* 2003.
74. Ledbetter C.A., Ramming D.W. Apricot cv. Robada. Application no. 502,516. Plant patent No. 9,890 USA. 1997.
75. Legave J.M. Aspects of floral necrosis before flowering in Apricot. *Ann Amélior Plantes.* 1978; 28: 333-340.
76. Legave J.M., García G., Marco F. Some descriptive aspects of drops process of flower buds, or young flowers observed on apricot tree in south of France. *Acta Hort.* 1982; 121: 75-83.
77. Lillecrapp A.M., Wallwork M.A., Sedgley M. Female and male sterility cause low fruit set in a clone of the 'Trevatt' variety of apricot (*Prunus armeniaca* L.). *Sci. Hort.-Amsterdam.* 1999; 82: 255-263.
78. Martínez-Tellez J, Crossa-Raynaud P. Contribution à l'étude du processus de la fécondation chez trois espèces de *Prunus*: *P. persica* (L.) Batsch, *P. cerasifera* Ehrh, *P. mahaleb* L. grâce à l'utilisation de couples de variétés mâle-stériles et mâle-fertiles. *Agronomie.* 1982; 2:333-340.
79. Matton DP, Nass N, Clarke AE, Newbiggin E. Self-Incompatibility: How plants avoid illegitimate offspring. *P Natl Acad Sci USA.* 1994; 91:1992-1997.
80. Mau S.L., Raff J., Clarke A.E. Isolation and partial characterization of components of *Prunus avium* L. styles, including an antigenic glycoprotein associated with a self-incompatibility genotype. *Planta.* 1982; 156: 505-516.
81. Mehlenbacher S.A., Cociu V., Hough L.F. Apricots (*Prunus*). In: Genetic resources of temperate fruit and nut crops (Moore J.N., Ballington J.R., eds). International Society for Horticultural Science, Wageningen, 1991; 65-107.
82. Milatović D, Nikolić D. Analysis of self-(in) compatibility in apricot cultivars using fluorescence microscopy. *J. Hort. Sci. Biotech.* 2007; 82: 170-174.
83. Monet R., Bastard Y. Effets d'une température modérément élevée: 25°C, sur les bourgeons floraux du pêcher. *Physiol Vég.* 1971; 9: 209-226.
84. Newbiggin, E., Anderson, M.A., Clark, A.E. Gametophytic self-incompatibility systems. *Plant Cell.* 1993; 5: 1315-1324.
85. Nyéki J., Szabó Z. Cross-incompatibility in stone fruits. *Hortic. Sci.* 28, 23-31. OKIE W.R., WERNER D.J., 1996. Genetic influence on flower bud density in peach and nectarine exceeds that of environment. *HortScience.* 1995; 31: 1010-1012.
86. Ramming D.W., Tanner O. 'Castlebrite' apricot. *Hort. Science.* 1978; 13:485.
87. Rodrigo J., Herrero M. Effects of pre-blossom temperatures on flower development and fruit set in apricot. *Sci. Hort.-Amsterdam.* 2002; 92: 125-135.
88. Romero C., Vilanova S., Soriano J.M., Burgos L., Llacer G., Badenes M.L. Análisis de la estructura genómica del locus S en *Prunus armeniaca* L. Identificación y secuenciación de dos alelos S. *Actas de Horticultura.* 2003; 39:167-168.
89. Sage T.L., Bertin R.I., Williams E.G. Ovarian and other late-acting self-incompatibility systems. In: Genetic control of self-incompatibility and reproductive development in flowering plants (Williams E.G., Clarke A.E., Knox R.B., eds). Ed Kluwer, Dordrecht, 1994; 116-140.
90. Sakurai K, Brown SK, Weeden NF. Determining the self-incompatibility alleles of Japanese apple cultivars. *Hort. Science.* 1997; 32:1258-1259.
91. Sakurai K., Brown S.K., Weeden N.F. Selfincompatibility alleles of apple cultivars and advanced selections. *Hort. Science.* 2000; 35: 116-119.
92. Sanzol J., Herrero M. The «effective pollination period» in fruit trees. *Sci Hort.-Amsterdam.* 2001; 90:1-17.
93. Sanzol J., Rallo P., Herrero M. Asynchronous development of stigmatic receptivity in the pear (*Pyrus communis* Rosaceae) flower. *Am J Bot.* 2003; 90: 78-84.
94. Sassa H., Hirano H., Ikehashi H. Self-incompatibility-related RNases in styles of Japanese pear (*Pyrus serotina* Rehd). *Plant Cell Physiol.* 1992; 33: 811-814.
95. Sassa H, Hirano H, Ikehashi H. Identification and characterization of stylar glycoproteins associated with self-incompatibility genes of Japanese pear, *Pyrus serotina* Redh. *Mol Gen Genet.* 1993; 241:17-25. 240.
96. Burgos L, et al. *Span J Agric Res.* 2004; 2(2):227-241.
97. Sassa H, Mase N, Hirano H, Ikehashi H. Identification of self-incompatibility-related glycoproteins in styles of apple (*Malus x domestica*). *Theor Appl Genet.* 1994; 89:201-205.
98. Schultz JH. 1948. Self-incompatibility in apricots. *J Am Soc Hort. Sci* 51, 171-174.

99. Seth J.N., Kuksal R.P., 1977. A note on the causes of poor fruit setting in apricot variety Charmagz. *Progressive Horticulture* 9 (2), 39-42.
100. Sonneveld T, Robbins TP, Boskovic R, Tobutt KR. Cloning of six cherry self-incompatibility alleles and development of allele-specific PCR detection. *Theor Appl Genet* 2001; 102:1046-1055.
101. Szabó Z., Nyéki J., 1991. Blossoming, fructification and combination of apricot varieties. *Acta Hort* 293, 295-302.
102. Tamura M., Ushijima K., Sassa H., Hirano H., Tao R., Gradziel T.M., Dandekar A.M. Identification of self-incompatibility genotypes of almond by allele-specific PCR analysis. *Theor Appl Genet.* 2000; 101:344-349.
103. Tao R., Habu T., Namba A., Yamane H., Fuyuhiko F., Iwamoto K., Sugiura A. Inheritance of S-f-RNase in Japanese apricot (*Prunus mume*) and its relation to self-compatibility. *Theor Appl Genet.* 2002a; 105:222-228.
104. Tao R., Habu T., Yamane H., Sugiura A. Characterization and cDNA cloning for S-f-RNase, a molecular marker for self-compatibility, in Japanese apricot (*Prunus mume*). *J Jpn Soc Hortic Sci.* 2002b; 71: 595-600.
105. Tao R., Habu T., Yamane H., Sugiura A., Iwamoto K. Molecular markers for self-compatibility in Japanese apricot (*Prunus mume*). *HortScience.* 2000; 36:1121-1123.
106. Tao R., Yamane H., Sassa H., Mori H., Gradziel T.M., Dandekar A.M., Sugiura A. Identification of stelar RNases associated with gametophytic self incompatibility in almond (*Prunus dulcis*). *Plant Cell Physiol.* 1997; 38: 304-311.
107. Tao R., Yamane H., Sugiura A., Murayama H., Sassa H., Mori H. Molecular typing of S-alleles through identification, characterization and C-DNA cloning for S-RNases in sweet cherry. *J Am Soc Hortic Sci.* 1999; 124: 224-233.
108. Thompson M.M., Liu L.J. Temperature, fruit set and embryo sac development in 'Italian'prune. *J Am Soc Hortic Sci.* 1973; 98: 193-197.
109. Tomimoto Y, Nakazaki T, Ikehashi H, Ueno H, Hayashi R. Analysis of self-incompatibility-related ribonucleases (S- RNases) in two species of pears, *Pyrus communis* and *Pyrus ussuriensis*. *Sci Hortic-Amsterdam.* 1996; 66:159-167.
110. Uriu K. Effect of post-harvest soil moisture depletion on subsequent yield of apricots. *Proc Amer Soc Hortic Sci.* 1964; 84:93-97.
111. Vachun Z. Étude de quelques propriétés morphologiques et physiologiques du pollen d'abricotier. Germination et croissance des tubes polliniques a basses temperatures. *Acta Hort* 1981; 85:387-417.
112. Van Gastel, A.J.G. Mutability of the self-incompatibility locus and identification of the S-bearing chromosome in *Nicotiana glauca*. Doctoral Thesis. Wageningen, 1976.
113. Vilanova S, Romero C, Abernathy D, Burgos L, Llácer G, Abbott A, Badenes ML. Construction and application of a bacterial artificial chromosome (BAC) library of *Prunus armeniaca* L. for the identification of clones linked to the self-incompatibility locus. *Molecular Gen Genomics.* 2003; 269:685-691.
114. Viti R., Monteleone P., Guerriero R. Incompatibility in apricot (*Prunus armeniaca* L.): Methodological considerations. *J Hortic. Sci.* 1997; 72:961-970.
115. Wiersma PA, Wu Z, Zhou L, Hampson C, Kappel F. Identification of new self-incompatibility alleles in sweet cherry (*Prunus avium* L.) and clarification of incompatibility groups by PCR and sequencing analysis. *Theor Appl Genet.* 2001; 102:700-708.
116. Williams R.R. The effect of summer nitrogen applications on the quality of apple blossom. *J Hortic Sci.* 1965; 40: 31-41.
117. Williams R.R. Pollination studies in fruit trees: III. The effective pollination period for some apple and pear varieties. *Rep Long Ashton Res Stn.* 1966; 136-138.
118. Williams R.R. Techniques used in fruit set experiments. In: *Towards regulated cropping* (Williams R.R., Wilson D., eds). Ed Grower Books, London, 1970a; 57-61.
119. Williams R.R. Factors affecting pollination in fruit trees. In: *Physiology of tree crops* (Luckwill L.L., Cutting C.V., eds). Ed Academic Press, London, 1970b; 193-207.
120. Williams R.R. An analysis of fruit-set determinants in 1969. In: *Towards regulated cropping* (Williams R.R., Wilson D., eds). Ed Grower Books, London, 1970c; 11-22.
121. Williams R.R., Brain P., Church R.M., Flook V.A. Flower receptivity, pollen transfer and fruit set variations during a single flowering period of Cox's Orange Pippin apple. *J. Hortic. Sci.* 1984; 59: 337-347.
122. Yaegaki H., Shimada T., Moriguchi T., Hayama H., Haji T., Yamaguchi M. Molecular characterization of S-RNase genes and S-genotypes in the Japanese apricot (*Prunus mume* Sieb. et Zucc.). *Sex Plant Reprod.* 2001; 13: 251-257.
123. Yamane H., Tao R., Murayama H., Sugiura A. Determining the S-genotypes of several sweet cherry cultivars based on PCR-RFLP analysis. *J. Hortic. Sci. Biotech.* 2000; 75: 562-567.
124. Zuccherelli S., Tassinari P., Broothaerts W., Tartarini S., Dondini L., Sansavini S. S-allele characterization in self-incompatible pear (*Pyrus communis* L.). *Sex Plant Reprod.* 2002; 15:153-158.