

# The Appropriate Management of Honey Bee Colonies for Pollination of *Rosaceae* Fruit Trees in Warm Climates

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## ABSTRACT

Most of the *Rosaceae* fruit trees, such as apple, pear, plum, almond and cherry, exhibit full self-incompatibility. Therefore, their fruit production completely depends on cross pollination. The ultimate pollen carrier in *Rosaceae* is the honey bee. In the present review, different bee-hive management techniques for improving fruit set and yield are discussed. The main manipulations are of colony density and the timing and number of colony introductions. In pear, it was found that increasing the density from 2.5 colonies ha<sup>-1</sup> to 5 colonies ha<sup>-1</sup> in one introduction at 10% full bloom (FB), did not increase bee activity on the trees and did not improve fruit set and yield. However, introducing the colonies sequentially (1.25 colonies ha<sup>-1</sup> at 10% FB and 1.25 colonies ha<sup>-1</sup> at FB) increased bee activity and consequently improved fruit set and yield. In apple, the combination of both treatments: increasing the density to 2.5 colonies ha<sup>-1</sup> at 10% FB and a second introduction of 2.5 colonies ha<sup>-1</sup> at FB, for a total of 5 colonies ha<sup>-1</sup>, increased the number of bees tree<sup>-1</sup>, their mobility between the rows and the proportion of “topworkers” compared with “sideworkers”. As a result, fruit set and yield were enhanced. In Japanese plum, highest bee activity, fruit set and yield, were achieved when colonies were introduced at four different times (multiple introductions). Each introduction was of a density of 1.25 colonies ha<sup>-1</sup> at 10% FB, 50% FB, FB and FB+3 days (total of 5 colonies ha<sup>-1</sup>). For all three species, there was a positive and statistically significant correlation between the average number of bees tree<sup>-1</sup> and fruit set or yield. The optimum number of bees tree<sup>-1</sup> min<sup>-1</sup> at FB was 6-7 for pear, 7-8 for Japanese plum and 12-14 for apple.

**Keywords:** apple, colony, hive, honey bee, fruit set, pear, plum, pollination, yield

**Abbreviations:** FB, full bloom; HD, high density; MI, multiple introduction; MIHD, multiple introduction of high density; SI, sequential introduction; SIHD, sequential introduction of high density

**Definitions:** sideworkers: honey bees collecting nectar from the side of the flower, without contacting the anther and stigma; topworkers: honey bees visiting the flower from the top, thus contacting the anther or stigma.

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## INTRODUCTION

In Israel, *Rosaceae* fruit trees, such as apple (*Malus domestica*), pear (*Prunus communis*), plum (*Prunus salicina*), almond (*Prunus dulcis*) and cherry (*Prunus avium*) are grown mostly in the relatively cooler northern regions. However, the climate is sometimes not cold enough during the winter, which leads to low chilling units, especially in apple and cherry (Zur and Gur 2000; Naor *et al.* 2003). On the other hand, in summer it is usually too warm, which is a problem for the compatibility between rootstock and scion, especially in pear (Gur 2000). As a result of these climate conditions, the trees suffer from poor yields. Another important problem that contributes to the low yields is insufficient pollination and fertilization. *Rosaceae* carry the S-RNase mediated gametophytic self-incompatibility (GSI) system, which prevents self-fertilization (Schneider *et al.*

2001a, 2001b; Beppu *et al.* 2002, 2003; Sapir *et al.* 2004; Schneider *et al.* 2004; Zisovich *et al.* 2004a, 2004b; Zisovich *et al.* 2005; Goldway *et al.* 2007a, 2007b). Rejection occurs when pollen of the S-haplotype matches one of the two S-loci in the pistil (Goldway *et al.* 2007a). For example, a pistil carrying an Sa-haplotype inhibits the growth of Sa-pollen but not of pollen with other haplotypes (McCubbin and Kao 2000).

Thus, an incompatible cultivar couple (both cultivars having identical S-genotypes) does not set fruit, whereas semi-compatible (both cultivars share one of the two S-genotypes) or fully-compatible (the two cultivars carry different S-genotypes) do set fruit. However, it was shown that in sub-optimal conditions for pollination and fertilization, the potency of cross-fertilization between semi-compatible cultivars is lower than that of fully-compatible cultivars (Goldway *et al.* 1999; Schneider *et al.* 2005; Goldway

*et al.* 2007a, 2007b).

Self-incompatible plants wholly depend on the intensive mobility of a pollen carrier between themselves and a compatible cultivar that blooms at the same time (McGregor 1976; Free 1993; Benedek 1996; Delaplane and Mayer 2000; Webster 2002).

Honey bees (*Apis mellifera*) are the most important pollinators of these species due to their high demand for pollen and nectar and their hairy body, which collects and disperses the pollen (McGregor 1976; Free 1993; Benedek 1996; Soltesz 1996; Delaplane and Mayer 2000; Stern *et al.* 2001, 2004). In addition, honey bee hives contain 30-50 thousand bees and are easy to mobilize. However, other bee species, such as *Osmia cornuta* (leafcutting bee) or *Bombus terrestris* (bumblebee) may also prove to be efficient pollinators. *O. cornuta* is more active than honey bees in lower temperatures (<15°C), makes better contact with the stigma and visits the flowers more frequently (Vicens and Bosch 2000; Maccagnani *et al.* 2003; Monzon *et al.* 2004). *B. terrestris* is also active at temperatures lower than 15°C (Lundberg 1980; Corbet 1993; Calzoni and Speranza 1996), carries more pollen grains, visits more flowers (Willmer *et al.* 1994), and deposits higher quantities of more compatible pollen grains per stigma than honey bees (Jacquemart *et al.* 2006). Nevertheless, both species are still not used commercially for pollination in orchards (Mayer *et al.* 1994; Mayer and Lunden 1997; Delaplane and Mayer 2000), and for the time being honey bees are the main pollinators.

Pome fruit, such as apple and pear, and stone fruit, such as plum, almond, apricot and cherry, are usually grown in temperate zones. Weather conditions during the blooming period may be unfavourable for flight of pollinating insects. Honey bees, in particular, are less active during cold, cloudy, rainy, and windy weather (McGregor 1976; Free 1993; Benedek 1996). Cool temperatures also affect pollen-tube growth and fertilization (Soltesz 1996; Westwood 1993). Consequently, low cross-pollination levels are usually one of the most yield-limiting factors (Dennis 1979; Free 1993; Hoopingarner and Waller 1993; Westwood 1993; Soltesz 1996; Dennis 2003).

In all *Rosaceae* fruit trees, the honey bee often abandons the target flowers in favor of flowers of fruit trees such as *Citrus* (Free 1993) and lychee (Stern and Gazit 1996), or competing flora that is more attractive and more rewarding for the honey bees (Free *et al.* 1960; Free and Spencer-Booth 1963; Dennis 1979; Delaplane and Mayer 2000). Moreover, honey bees tend to restrict their mobility to one row, which usually contains a single cultivar (Williams and Smith 1967; Eisikowitch *et al.* 1999).

In addition, the “effective pollination period” (EPP), i.e. ovule longevity minus the time between pollination and fertilization, is very short in apple and pear (Williams 1966). In ‘Bartlet’ pear it could last only 1-2 days from anthesis at 9-10°C (Lombard *et al.* 1971), whereas in ‘Red-Delicious’ apple it could be even shorter (Dennis 1979, 1986). Thus, although the stigma remains receptive for longer periods of time, pollination needs to be accomplished in 1-2 days for fertilization to occur before ovule degeneration. Therefore, any technique that would increase the activity of bees and their efficiency in cross-pollination should improve the yield.

This review describes the application of some honey bee management procedures aimed at improving yields, which have been tested in three species of *Rosaceae* fruit trees – pear and apple (from the pome group), and plum (from the stone group).

## PEAR (*Pyrus communis*)

Pear flowers are not attractive for honey bees (McGregor 1976; Mayer 1994; Mayer and Lunden 1997) due to the low volume of nectar secreted from the flowers (3 µl or less) and its low sugar concentration (<20% w/v) (McGregor 1976; Free 1993; Delaplane and Mayer 2000; Farkas *et al.*

2002), in contrast to higher sugar concentrations of cherry (24%), peach (29%) and apple (46%) (Vansell 1946; Free 1993; Delaplane and Mayer 2000). Moreover, the amount and quality of the nectar is highly variable since its production is affected by weather conditions (Benedek *et al.* 2000). Yet, this low nectar attractiveness is partly compensated by the abundance of pollen grains (1.2 mg flower<sup>-1</sup>), (McGregor 1976; Free 1993; Delaplane and Mayer 2000). Furthermore, foragers that collect pollen collect fresh and viable pollen, compared to “old” pollen that sticks to a forager that collects nectar (Kendall 1973; Klugness *et al.* 1983), and carry more pollen grains on their body hair than nectar collectors (Free and Williams 1972). Pollen-gatherers also make better contact with the stigma, further increasing their efficiency as pollinators (Vicens and Bosch 2000; Monzon *et al.* 2004). As a result, pear trees need fewer bees for pollination compared with apple or cherry (Delaplane and Mayer 2000).

Mayer *et al.* (1990) stated that 10-15 bees tree<sup>-1</sup> min<sup>-1</sup> are required for adequate pollination of pear. In the north-west of the U.S.A, the recommended density of colonies for pear orchard to reach this number of bees per tree ranges between a low density of one colony ha<sup>-1</sup> and a high density of 5 colonies ha<sup>-1</sup> (Humphry-Baker 1975; Levin 1986; Mayer *et al.* 1986; Kevan 1988; Williams 1994; Scott-Dupree *et al.* 1995). In Israel, it is recommended to use 2.5 colonies ha<sup>-1</sup> (Dag *et al.* 2003).

The timing of colony introduction, in relation to the stage of blooming, strongly influences the number of bees that visit the trees. Many reports have shown that placing hives in the orchard before the main blooming has taken place, leads the bees to abandon the orchard in favour of competing flowers in the vicinity, to which they establish constancy (Mayer *et al.* 1986; Free 1993). Therefore, it was recommended to delay the placement of bee colonies in pear orchards until bloom reached 25-50% (Humphry-Baker 1975; Kevan 1988). Although the introduction of colonies at the right time exposes the bees to massive blooming in the target orchard, still they tend to widen their forage area gradually, and may even abandon the target orchard as a result (Free *et al.* 1960). To overcome this problem, Al Tikrity *et al.* (1972) suggested the introduction of additional colonies at a later date, thus the new bees are attracted at first to the pear flowers before discovering the competing bloom. This sequential introduction was first tried in pear orchards in Washington (Mayer 1994) and was found to raise the number of bees in the orchard (although only for 1 day) and the consequent fruit set. The yields in this study were not recorded. However in cranberry, Shimanuki *et al.* (1967) found no advantage in placing honey bee colonies before rather than during peak bloom.

Encouraged by the positive results from the preliminary studies of Mayer (1994) in the temperate climate of Washington, Stern *et al.* (2004) established a large scale 4-year experiment in the hot climate of Israel (*ca.* 38°C max with <35% RH during the summer – May till October, and 2°C min with annual precipitation of about 500 mm during the winter – December till February). The aim of their study was to evaluate the effect of doubling the density of colonies from the recommended use of 2.5 ha<sup>-1</sup> to 5 colonies ha<sup>-1</sup> (high density treatment = HD) and of sequential introduction (SI) of colonies (half at 10% full bloom (FB) and half at FB) with or without increasing density on honey bee activity and on their effectiveness as pollinators of ‘Spadona’, the principle pear cultivar in Israel, and the consequences for fruit set and yield.

They found that increasing colony density to 5 colonies ha<sup>-1</sup> (HD) usually did not improve bee activity and did not increase pollination, fruit set and yield. However, introduction of new “naïve” bees to the orchard at FB in the SI treatment, increased bee activity and mobility between rows. Since the second introduction in the SI treatment was at the most important stage of the bloom, i.e. the peak, a low colony density of a total of only 2.5 colonies ha<sup>-1</sup> was sufficient for achieving agreeable fruit set.

Further support for the effect of the SI treatment came from analyzing the proportion of pear pollen pellets in pollen traps placed at the colony entrance. The average proportion of pear pollen pellets was significantly higher in the SI treatment (35%) compared to the HD and control (16%) treatments. Thus, the SI treatment increased the colony's relative allocation of pollen foragers to pear.

As a result of a general increase in bee activity and mobility between rows, and specifically of pear pollen foragers, fruit set and yield increased significantly, and sometimes also the number of seeds in the fruit increased, leading to larger fruit.

The significant positive correlation between the number of bees tree<sup>-1</sup> and yield, indicates, as previously found in five other pear cultivars (Stephen 1958), a strong relationship between bee activity and yield.

However, whereas the usual recommendation in the USA for optimum yield of mature pears is 10-15 bees tree<sup>-1</sup> min<sup>-1</sup> (Mayer *et al.* 1990), Stern *et al.* (2004) found in the temperate climate of Israel that 6-7 bees tree<sup>-1</sup> min<sup>-1</sup> sufficed.

It seems that the main reason for the relatively low bee activity on pear is the competing blooms surrounding the pear orchards in Israel. The pear blooming season in Israel (March-April) is within the peak blooming season of wild flowers (Zohary 1962), whereas in other countries with temperate climates, which grow 'Spadona', the pear blooming season coincides with only the beginning of the flowering of wild flowers. Thus, in Israel, a high proportion of bees forage outside of the orchard in the open fields.

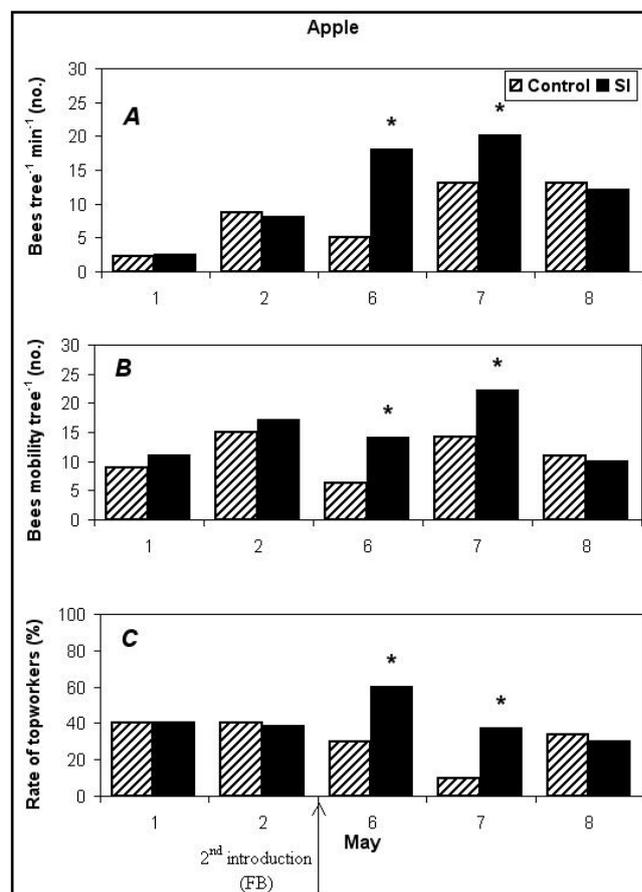
#### APPLE (*Malus domestica*)

Apples are more attractive to honey bees than pears, mainly because of their nectar (McGregor 1976; Free 1993; Delaplane and Mayer 2000). Nectar concentration in apple flowers is much higher than in pear flowers at the same time in the morning hours (>45% and <20% in apple and pear, respectively) (Vanzell 1946; Schneider *et al.* 2002; Zisovich 2003; Schneider *et al.* 2004) and in some cultivars, especially those of the 'Delicious' cultivars, it is very easy to collect (Dennis 1979, 1986, 2003; Schneider *et al.* 2004). However, although pollen collectors are better pollinators than nectar collectors also in apple (Dag *et al.* 2005) the pollination efficiency in apple is much lower than in pear, which is especially attractive for bees that collect pollen (Free 1993; Delaplane and Mayer 2000; Zisovich 2003).

In addition, the flower structure of some apple cultivars, such as 'Delicious', reduce pollination efficiency. On these flowers there are gaps at the base of the stamens that enable "sideworking" honey bees to obtain nectar without contacting the anthers and stigmas of the blossom (Roberts 1945). Robinson (1979) found that the gap that facilitates "sideworking" in 'Delicious' was considerably greater than in other cultivars. As a result, this cultivar is particularly susceptible to "sideworking". This has since been confirmed by Dennis (1979), Robinson (1979), Kuhn and Ambrose (1982), Mayer (1984), DeGrandi-Hoffman *et al.* (1985), Free (1993) Westwood (1993), Tomson and Goodell (2001) and Schneider *et al.* (2002, 2004). As a result, the pollination rate in 'Delicious' apple is particularly low compared with other cultivars.

Robinson and Fell (1981) found that after a single honey bee visit to a 'Delicious' flower, fruit-set was 8% when the visit was by a "sideworker", compared to 50% when the visit was by a "topworker". Collecting nectar from the side, without touching the anthers and stigmas, is very convenient for the bee, but requires time to learn (DeGrandi-Hoffman *et al.* 1985).

Schneider *et al.* (2004) found that 'Golden Delicious' also has wide gaps at the base of the stamens. However, more than double the number of "topworkers" were found in 'Golden Delicious' compared to 'Delicious', due to the wider diameter of the stamens filament spread of 'Golden Delicious' flowers. This facilitates nectar collection from the top and thus increases pollination effectiveness, leading



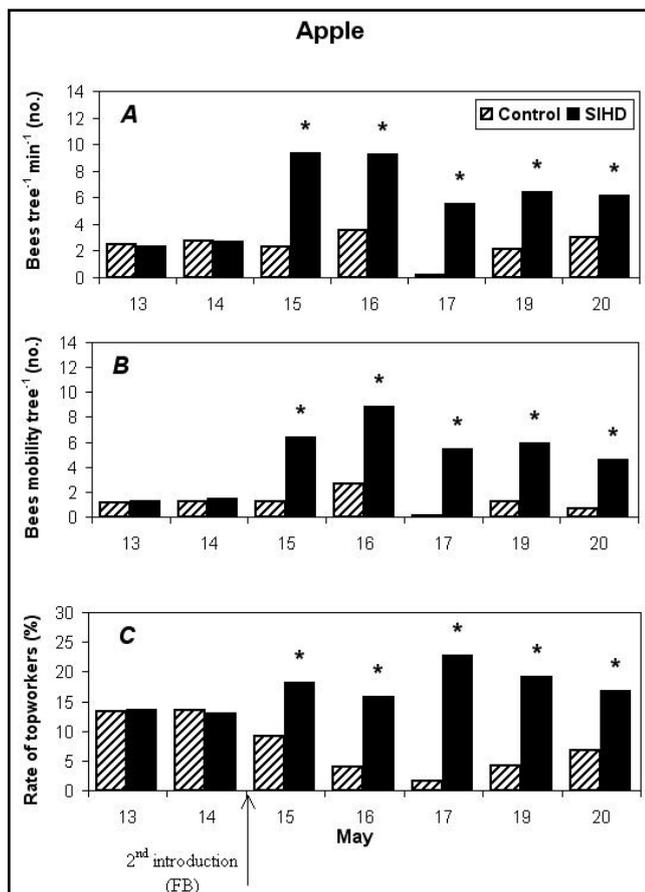
**Fig. 1** Effect of sequential introduction (SI) of colonies on the average number of bees per tree (A), mobility between rows (B) and proportion of "topworkers" (C) during the flowering period of 'Delicious' apple. Both treatments had a total of 2.5 colonies per ha (the SI treatment started with 1.25 colonies per ha). \* = Separation between treatment means on each day by Duncan's new multiple range test,  $P = 0.05$ . (Reproduced from Stern *et al.* (2001) *Journal of Horticultural Science and Biotechnology* 76, 17-23, with kind permission).

to higher fruit set and yield compared to 'Delicious'.

To overcome the low efficiency pollination of 'Delicious', Mayer *et al.* (1986) stated that 20-25 bees tree<sup>-1</sup> min<sup>-1</sup> are required for adequate pollination of 'Delicious' apple compared to only 10-15 bees for pollination of pear (Mayer *et al.* 1990). For such high bee activity, it is recommended in the USA, to place colonies containing 4-6 brood combs at a density of 2.5 colonies ha<sup>-1</sup> in apple orchards (Mayer *et al.* 1986; Hoopingarner and Waller 1993). In Israel it is recommended to use 2.5 colonies ha<sup>-1</sup> with seven brood combs (Ben-Porat *et al.* 1997; Dag *et al.* 1999, 2003). However the yields were still low, especially of 'Delicious' apple.

Encouraged by the positive results from the preliminary studies on pears, Stern *et al.* (2001) established a large scale 4-year experiment in Israel. As was previously examined on pear, the aim of this study was to evaluate the effects of doubling the density of colonies and of sequential introduction of colonies (with or without increasing density) on honey bee activity and on their effectiveness as pollinators and the consequences for fruit set and yield. The experiments were carried out with 'Delicious', the principle apple cultivar in Israel.

As in pears, it was found that sequential bee hive introduction (SI), even in low ratio of 2.5 colonies ha<sup>-1</sup>, improved the number of bees tree<sup>-1</sup> and their mobility between the rows (Fig. 1A, 1B). Furthermore, although there were just half the number of colonies in the SI treatment compared to the control at the beginning of the blooming period, there was no difference in bee foraging activity level between both treatments, probably because there were still only few open flowers at that stage. In addition, the proportion of "topworkers" was considerably higher in SI



**Fig. 2** Effect of sequential introduction of high density colonies (SIHD) on the average number of bees per tree (A) mobility between rows (B) and proportion of “topworkers” (C) during the flowering period of ‘Delicious’ apple. The SIHD treatment had 5.0 colonies per ha (initially, 2.5 colonies per ha) and the control had 2.5 colonies per ha. \* = Separation between treatment means on each day by Duncan’s new multiple range test,  $P = 0.05$ . (Reproduced from Stern *et al.* (2001) *Journal of Horticultural Science and Biotechnology* 76, 17-23, with kind permission).

compared to the control treatment (Fig. 1C). These effects were observed during only 2 days of FB, but led to increased cross-pollination and apple pollen collection (assessed with pollen traps), and resulted in greater fruit set and yield (by 50-80%).

It should be noted that in contrast to pear, in which the high density (HD) treatment of 5.0 colonies ha<sup>-1</sup> was not effective, in apple both HD and SI treatments conferred similar fruit set enhancement. In both pear and apple, the SI treatment was effective with only a total of 2.5 colonies ha<sup>-1</sup>.

Following the observation that doubling the colony density and sequential introduction of colonies increased bee pollination activity a treatment that combined the two techniques was applied. It was assumed that better pollination efficiency could be achieved by extending the effect of the treatments on bee activity for longer than just 2 days (Stern *et al.* 2001). Indeed, the combination of the two treatments (SIHD = Sequential Introduction of High Density colonies) significantly increased bee activity in trees, and their mobility between the rows from the second introduction at FB until the end of the blooming period (Fig. 2). There was a strong positive correlation between these two parameters ( $R^2 = 0.90$ ) as found also in avocado (Vitanage 1990; Ish-Am and Eisikowitch 1998) and almond (Eisikowitch 1999). The SIHD treatment also enhanced the bees’ efficiency as pollinators by considerably increasing the proportion of “topworkers”. The high proportion of “topworkers” soon after the introduction of new colonies is explained by the fact that “sideworking” is a learned behaviour that most naïve bees have not acquired. DeGrandi-Hoffman *et al.* (1985) stated that the percentage

of sideworking honey bees steadily increased from 21% on the first day of ‘Delicious’ bloom to 60% on the fifth day. This change in behaviour seems to be due to “sideworkers” spending less time than “topworkers” in reaching the nectararies (Robinson 1979).

The combined effects of the SIHD treatment in the Stern *et al.* (2001) study, resulted in almost double the fruit set compared to the control. There was a positive correlation between the average number of bees tree<sup>-1</sup> and fruit set and yield. They proposed 12-14 bees tree<sup>-1</sup> min<sup>-1</sup> at FB for optimum yield. At these numbers there are enough bees which working as a “topworker” and moving from row to row.

In summary, we can conclude from the experiments in ‘Delicious’ apple, that: 1) A large number of foragers tree<sup>-1</sup> increases the pollination level. 2) High bee mobility between rows increases the cross-pollination level, and 3) A high proportion of “topworkers” increases pollination efficiency. All these factors were expressed in higher fruit set and yields (by 50-100%). Thus, it is recommended to introduce the colonies sequentially (half at 10% FB and half at FB) at a final ratio of 5 colonies ha<sup>-1</sup> in order to reach 12-14 bees tree<sup>-1</sup> min<sup>-1</sup> at FB.

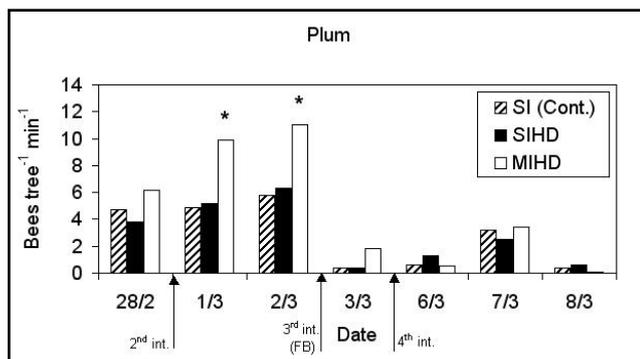
### JAPANESE PLUM (*Prunus salicina*)

The main reason for low fertility of Japanese plum in the Mediterranean region and especially in Israel is the very early bloom, from mid February until mid March. During this period it is usually cold, windy and rainy. This climate is unfavourable for bee flight, pollination, pollen-tube growth and fertilization (McGregor 1976; Free 1993; Westwood 1993; Benedek 1996; Delaplane and Mayer 2000). Still, the recommended density of colonies for plum orchard is not always high, and ranges between 2.5 (McGregor 1976; Crane and Walker 1984; Kevan 1988; Scott-Dupree *et al.* 1995) and 5 colonies ha<sup>-1</sup> (Standifer and McGregor 1977; Mayer *et al.* 1986). Yet, the activity of bees in the orchard is not satisfactory (Mayer *et al.* 1986), leading to low yields. Another obstacle is that Japanese plum cultivars do not flower synchronously. The early flowering cultivars bloom in mid February, whereas the late ones bloom in the end of April (Westwood 1993).

The ‘Black-Diamond’<sup>®</sup> cultivar which was bred by Sun-World Corp. and registered also as Suplumeleven<sup>™</sup>, has become, in the last decade, one of the most important plum cultivars in Israel, Europe and the USA. In our effort to find suitable pollinizers for ‘Black-Diamond’, with overlapping flowering, we found only semi-compatible cultivars, one of which is ‘Angeleno’ (Sapir *et al.* unpublished). In addition, ‘Black-Diamond’ and ‘Angeleno’ bloom during the first half of March, when climate conditions are unfavourable (see above). Therefore, the possibility of pollen deficiency is very high, and also in this case it was proposed by Mayer *et al.* (1986) and Standifer and McGregor (1977) to enhance bee activity in order to increase yield. The treatment applied was a combination of multiple introductions (every 2-3 days) and a high density of colonies (Sapir *et al.* unpublished).

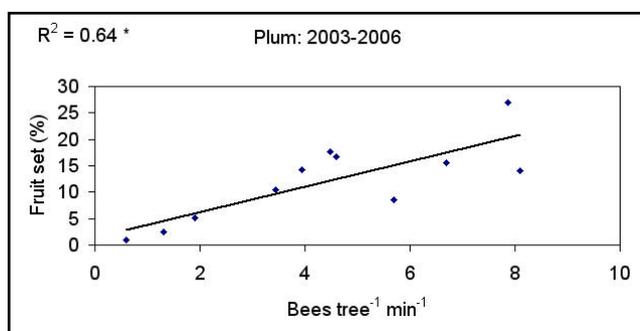
At the beginning of this study it was found that SI of colonies at a low rate of 1.25 + 1.25 colonies ha<sup>-1</sup> (at 10% FB and at FB), compared to the control treatment of the same density but with one introduction at 10% FB, significantly enhanced bee activity, from 5.7 to 8.1 bees tree<sup>-1</sup> min<sup>-1</sup>. Consequently, fruit set increased from 8.5 to 14.1%, and yield increased from 64 to 84 kg tree<sup>-1</sup>. The results of the SI treatment were similar to those of a high density treatment of 5 colonies ha<sup>-1</sup>, but with only half the number of colonies.

Following these findings Sapir *et al.* (unpublished data) tried to combine the SI treatment with HD treatment (SIHD) to test whether HD with a final total of 5 colonies ha<sup>-1</sup> (SIHD) confers a further advantage to sequential introduction with a final total of only 2.5 colonies ha<sup>-1</sup> (SI=control). They also tested whether four multiple introductions, every two-three days, with a total of 5 colonies ha<sup>-1</sup> (MIHD), is



**Fig. 3** Effect of sequential introduction of high density colonies (SIHD) and multiple introduction of high density colonies (MIHD), on the average number of bees tree<sup>-1</sup> min<sup>-1</sup> during the flowering period of 'Black-Diamond'® Japanese plum. The SI (control) treatment had 1.25 colonies ha<sup>-1</sup> at 10% FB and 1.25 colonies ha<sup>-1</sup> at FB (total of 2.5 colonies ha<sup>-1</sup>). The SIHD treatment had 2.5 colonies ha<sup>-1</sup> at 10% FB and 2.5 colonies ha<sup>-1</sup> at FB (total of 5 colonies ha<sup>-1</sup>). The MIHD treatment had 1.25 colonies ha<sup>-1</sup> at each of four introductions – 10% FB, 50% FB, FB and FB + 3 days (total of 5 colonies ha<sup>-1</sup>).

\* Separation between treatment means (MIHD versus SI and SIHD) in each day by Duncan's new multiple range test,  $P = 0.05$ .



**Fig. 4** Relationship between the average number of honey bees tree<sup>-1</sup> and the percentage of initial fruit set six weeks after FB in all treatments (3 treatment year<sup>-1</sup> X 4 years – 2003-2006). In 2005, one record (MI) was not included due to a heavy yield that demanded strong thinning. \* Significant at  $P = 0.05$ .

better than the SI treatment (only two introductions, for a total of 2.5 colonies ha<sup>-1</sup>).

They found that the MIHD treatment, combining sequential introduction with high colony density, increased dramatically the number of bees tree<sup>-1</sup> (Fig. 3). This high bee activity was observed especially during the first half of the flowering period (March 1-2), which included the two days following the second introduction, just prior to FB, until the "winter climate" began (March 3). This combined treatment (MIHD) significantly increased fruit set (20%) of 'Black Diamond' similarly the fruit set of the adjacent cultivar 'Angeleno' was also significantly increased by 36%.

Combining the 4 years of the study, there is a strong positive correlation between the average number of honey bees tree<sup>-1</sup> min<sup>-1</sup> and initial fruit set (Fig. 4). Higher percentage of fruit set led to higher yields. Since 20% fruit set led to the optimum yield of 70 kg tree<sup>-1</sup> we can conclude that at least 7-8 bees tree<sup>-1</sup> are required to achieve high yields in Japanese plum.

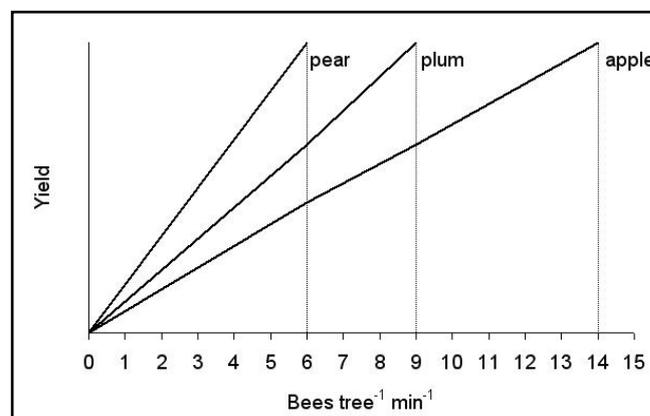
## CONCLUDING REMARKS

We found a significant positive correlation in pear, apple and Japanese plum, between the number of bees tree<sup>-1</sup> and fruit set percentage; high fruit set percentage leads to high yields. Hence, honey bees are very important for improving cross-pollination, especially in species with self-incompatible cultivars, and especially when only semi-compatible cultivars are present.

However, for each species there is a different optimum of bee number in order to reach maximum yield. In pear it is 6-7 bees tree<sup>-1</sup> min<sup>-1</sup>, in Japanese plum 7-8 bees tree<sup>-1</sup> min<sup>-1</sup> and in apple 12-14 bees tree<sup>-1</sup> min<sup>-1</sup> (Fig. 5). It seems that the number of bees required is affected by the attractiveness of the flower, pollination efficiency, genetic compatibility between cultivars and more.

In order to achieve these numbers of bees the colonies have to be managed properly. In all cases it was found that to get the best efficiency the colonies had to be introduced sequentially – 2 times along the blooming period in pear and apple and even 4 times in Japanese plum. Based on the US and Israeli experience, for areas with similar climates (e.g., Australia, South Africa, and southern Europe) we strongly recommend multiple introductions for all *Rosaceae* species, since these introduce naïve bees to the orchard every 2-3 days, thus improving bee activity level on the target trees and pollination efficiency. Consequently fruit set and yields increase.

The combination of this technique together with the total number of colonies, are very important and have to be learned for each species. To conclude, sequential introduction instead of a single introduction at the beginning of flowering, can improve pollination efficiency with a reduction in bee hive costs.



**Fig. 5** Schematic relationship found between the average number of honey bees tree<sup>-1</sup> and the optimal yield in each of the three crops – pear, plum and apple.

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## REFERENCES

- Al-Tikrity WS, Benton AW, Risius ML, Clarke WW (1972) The effect of length of stay of honey bee colony in a crownvetch field on its foraging behaviour. *Journal of Apicultural Research* 11, 51-57
- Benedek P (1996) Insect pollination of fruit crops. In: Nyeki J, Soltesz M (Eds) *Floral Biology of Temperate Zone Fruit Trees and Small Fruits*, Akademiai Kiado, Budapest, Hungary, pp 287-340
- Benedek P, Kocsis-Molnar G, Nyeki J (2000) Nectar production of pear (*Pyrus communis* L.) cultivars. *International Journal of Horticultural Science* 6, 67-75
- Ben-Porat A, Doron I, Dag A (1997) Apple pollination. *Alon Hanotea* 51, 76-78 (in Hebrew)
- Beppu K, Takemoto Y, Yamane H, Yaegaki H, Yamaguchi M, Kataoka I, Tao R (2003) determination of S-haplotypes of Japanese plum (*Prunus salicina* Lindl.) cultivars by PCR and cross-pollination tests. *Journal of Horticultural Science and Biotechnology* 78, 315-318
- Beppu K, Yamane H, Yaegaki H, Yamaguchi M, Kataoka I, Tao R (2002) Diversity of S-RNase genes and S-haplotypes in Japanese plum (*Prunus salicina* Lindl.). *Journal of Horticultural Science and Biotechnology* 77, 658-665

- Calzoni GL, Speranza A** (1996) Pear and plum pollination: honey bees, bumble bees or both? *Acta Horticulturae* **423**, 83-90
- Corbet SA** (1993) Wild bees for pollination in the agricultural landscape. In: Bruneau E (Ed) *Bees for Pollination*, Commission of the European Communities, Brussels, Belgium, pp 175-189
- Crane E, Walker P** (1984) *Pollination Directory for World Crops*, International Bee Research Association, London
- Dag A, Doron I, Stern RA** (2003) Recommendation for pollination of apple, pear and plum, 2003. Newsletter – *The Extension Service, Galilee-Golan Region* (in Hebrew), 2 pp
- Dag A, Stern RA, Doron I** (1999) Pollination and fertilization in apple. *Alon Hanotea* **53**, 114-116 (in Hebrew)
- Dag A, Stern RA, Shafir S** (2005) Honey bee (*Apis mellifera*) strains differ in apple (*Malus domestica*) pollen foraging preference. *Journal of Apicultural Research* **44**, 15-20
- Degrandi-Hoffman G, Hoopingartner R, Baker KK** (1985) The influence of honey bee 'Sideworking' behavior on cross-pollination and fruit set in apple. *HortScience* **20**, 397-399
- Delaplane KS, Mayer DF** (2000) *Crop Pollination by Bees*, CABI Publishing, Wallingford, UK
- Dennis FG Jr** (1979) Factors affecting yield in apple with the emphasis on 'Delicious'. *Horticultural Reviews* **1**, 395-422
- Dennis FG Jr** (1986) Apple. In: Monselise SP (Ed) *Handbook of Fruit Set and Development*, CRC Press, Boca Raton, FL, USA, pp 1-46
- Dennis FG Jr** (2003) Flowering, pollination and fruit set and development. In: Ferre DC, Warrington IJ (Eds) *Apple: Botany, Production and Uses*, CABI Publishing, Wallingford, UK, pp 153-164
- Eisikowitch D, Loper G, DeGrandi-Hoffman G** (1999) Honey bees movement among trees in the almond orchard in Israel. *Proceedings of the 36<sup>th</sup> Apimondia Congress*, Vancouver, Canada, pp 2-17
- Farkas A, Orosz-Kovacs Z, Szabo L** (2002) Insect attraction of flowers in pear cultivars. *Acta Horticulturae* **596**, 773-776
- Free JB** (1993) *Insect Pollination of Crops* (2<sup>nd</sup> Edn), Academic Press, London
- Free JB, Free NW, Jay SC** (1960) The effect on foraging behaviour of moving honey bee colonies to crops before or after flowering has begun. *Journal of Economic Entomology* **53**, 564-566
- Free JB, Spencer-Booth Y** (1963) The foraging areas of honey bee colonies in fruit orchards. *Journal of Horticultural Science* **38**, 129-137
- Free JB, Williams IH** (1972) The transport of pollen on the body hair of honeybees and bumblebees. *Journal of Applied Ecology* **9**, 609-615
- Goldway M, Shai O, Yehuda H, Matityahu A, Stern RA** (1999) 'Jonathan' apple is a lower-potency pollenizer of 'Topred' than 'Golden Delicious' due to partial S-allele incompatibility. *Journal of Horticultural Science and Biotechnology* **74**, 381-385
- Goldway M, Sapir G, Stern RA** (2007a) Molecular basis and horticultural application of the gametophytic self-incompatibility system in *Rosaceae* tree fruits. In: Janick J (Ed) *Plant Breeding Reviews* **28** (Chapter 7), John Wiley and Sons, Inc. Publishing, USA, pp 215-237
- Goldway M, Sapir G, Raz A, Stern RA** (2007b) The molecular understanding of the S-RNase-mediated gametophytic self-incompatibility system and its impact on the cultivation of *Rosaceae* fruit trees. *Advances in Plant Physiology* **10**, in press
- Gur A** (2000) Effects of high temperatures at the root zone and the graft union on the development of temperate fruit trees. In: Erez A (Ed) *Temperate Fruit Crops in Warm Climates*, Kluwer Academic Publishers, Dordrecht, Netherlands, pp 123-135
- Hoopingartner RA, Waller GD** (1993) Crop pollination. In: Graham JM (Ed) *The Hive and the Honeybee*, Dadant and Sons, Carthage, Illinois, USA, pp 1044-1080
- Humphry-Baker P** (1975) *Pollination and Fruit Set in Tree Fruits*, British Columbia Department of Agriculture, Victoria
- Ish-Am G, Eisikowitch D** (1998) Mobility of honey bees (*Apidae*, *Apis mellifera* L.) during foraging in avocado orchards. *Apidologie* **29**, 209-219
- Jacquemart AL, Michotte-van der Aa A, Raspe Q** (2006) Compatibility and pollinator efficiency tests on *Pyrus communis* L. cv. 'Conference'. *Journal of Horticultural Science and Biotechnology* **81**, 827-830
- Kendall DA** (1973) Viability and compatibility of pollen on insects visiting apple blossom. *Journal of Applied Ecology* **10**, 847-853
- Kevan PG** (1988) *Pollination: Crops and Bees*, Ontario Ministry of Agriculture and Food, Publication No. 72
- Klugness M, Thorp R, Briggs D** (1983) Field testing the germinability of almonds pollen. *Journal of Horticultural Science* **58**, 229-235
- Kuhn ED, Ambrose T** (1982) Foraging behaviour of honey bees on 'Golden Delicious' and Delicious Apple. *Journal of the American Society for Horticultural Science* **107**, 391-395
- Levin MD** (1986) *Using Honey Bees to Pollinate Crops*, US Department of Agriculture, Leaflet 549
- Lombard P, Cordy C, Hansen E** (1971) Relation of postbloom temperatures to 'Bartlett' pear maturation. *Journal of the American Society for Horticultural Science* **96**, 799-801
- Lundberg H** (1980) Effects of weather on foraging-flights of bumblebees (*Hymenoptera*, *Apidae*) in a subalpine/alpine area. *Holarctic Ecology* **3**, 104-110
- Maccagnani B, Ladurner E, Santi F, Burgio G** (2003) *Osmia cornuta* (*Hymenoptera*, *Megachilidae*) as pollinator of pear (*Pyrus communis*): fruit-and seed-set. *Apidologie* **34**, 207-216
- Mayer DF** (1984) Behavior of pollinators on *Malus*. *Fifth International Pollination Symposium*, Versailles **1983**, 387-390
- Mayer DF** (1994) Sequential introduction of honey bee colonies for pear pollination. *Acta Horticulturae* **367**, 267-269
- Mayer DF, Johansen CA, Burgett DM** (1986) *Bee Pollination of Tree Fruits*, Washington State University, Pacific Northwest Cooperative Extension Bulletin. No. **0282**
- Mayer DF, Lunden JD** (1997) Comparison of commercially managed bumblebees and honeybees (*Hymenoptera: Apidae*) for pollination of pears. *Acta Horticulturae* **437**, 283-287
- Mayer DF, Miliczky ER, Lunden JD** (1990) Pollination of pears. In: *Pear Production in the Pacific Northwest*, University of California Press, Davis, CA
- Mayer DF, Patten KD, McFarlane RP** (1994) Pear pollination with managed bumblebee (*Hymenoptera: Apidae*) colonies. *Melandria* **50**, 20-23
- McCubbin, AG, Kao TH** (2000) Molecular recognition and response in pollen and pistil interaction. *Annual Review of Cell and Developmental Biology* **16**, 333-364
- McGregor SE** (1976) *Insect Pollination of Cultivated Crop Plants*, US Department of Agriculture Handbook No. **496**
- Monzon VH, Bosch J, Retana J** (2004) Foraging behavior and pollinating efficiency of *Osmia cornuta* (*Hymenoptera: Megachilidae*) and *Apis mellifera* (*Hymenoptera: Apidae*) on 'Comice' pear. *Apidologie* **35**, 575-585
- Naor A, Flaishman M, Stern RA, Aharon M, Erez A** (2003) Temperature effects on dormancy completion of vegetative buds in apple. *Journal of the American Society for Horticultural Science* **128**, 636-641
- Roberts RH** (1945) Blossom structure and setting of 'Delicious' and other apple varieties. *Proceedings of the American Society for Horticultural Science* **46**, 87-90
- Robinson WS** (1979) Effect of apple cultivar on foraging behaviour and pollen transfer by honey bees. *Journal of the American Society for Horticultural Science* **104**, 596-598
- Robinson WS, Fell RD** (1981) Effect of honey bee foraging behaviors on 'Delicious' apple set. *HortScience* **16**, 326-328
- Sapir G, Stern RA, Eisikowitch D, Goldway M** (2004) Cloning of four new Japanese plum S-allele and determination of the compatibility between cultivars by PCR analysis. *Journal of Horticultural Science and Biotechnology* **79**, 223-227
- Schneider D, Eisikowitch D, Goldway M, Stern RA** (2004) A comparative study of the superior fertility of 'Smoothy Golden Delicious' apple *Journal of Horticultural Science and Biotechnology* **79**, 596-601
- Schneider D, Stern RA, Eisikowitch D, Goldway M** (2001a) Analysis of S-allele by PCR for determination of compatibility in the Red Delicious apple orchard. *Journal of Horticultural Science and Biotechnology* **76**, 596-600
- Schneider D, Stern RA, Eisikowitch D, Goldway M** (2001b) Determination of the self fertilization potency of Golden Delicious apple. *Journal of Horticultural Science and Biotechnology* **76**, 259-263
- Schneider D, Stern RA, Eisikowitch D, Goldway M** (2002) The relationship between floral structure and honeybee pollination efficiency in Jonathan and Topred apple cultivars. *Journal of Horticultural Science and Biotechnology* **77**, 48-51
- Schneider D, Stern RA, Goldway M** (2005) A comparison between semi- and fully-compatible apple pollinators growth under suboptimal pollination condition. *HortScience* **40**, 1280-1282
- Scott-Dupree C, Winston M, Hergert G, Jay SC, Nelson D, Gates J, Termeer B, Otis G** (Eds) (1995) *A Guide to Managing Bees for Crop Pollination*, Canadian Association of Professional Apiculturalists, British Columbia, Canada
- Shimanuki H, Lehnert T, Stricker M** (1967) Differential collection of cranberry pollen by honey bees. *Journal of Economic Entomology* **60**, 1031-1033
- Soltész M** (1996) Requirements for successful fruit set in orchards. In: Nyeki J, Soltész M (Eds) *Floral Biology of Temperate Zone Fruit Trees and Small Fruits*, Akademiai Kiado, Budapest, Hungary, pp 257-286
- Standifer L, McGregor SE** (1977) Using honey bees to pollinate crops. *US Department of Agriculture*, Leaflet No. **549**
- Stephen WP** (1958) Pear pollination studies in Oregon. *Oregon Agricultural Experimental Station Bulletin* No. **43**
- Stern RA, Eisikowitch D, Dag A** (2001) Sequential introduction of honeybee colonies and doubling their density increase cross-pollination, fruit set and yield in 'Red Delicious' apple. *Journal of Horticultural Science and Biotechnology* **76**, 17-23
- Stern RA, Gazit S** (1996) Lychee pollination by the honeybee. *Journal of the American Society for Horticultural Science* **121**, 152-157
- Stern RA, Goldway M, Zisovich AH, Shafir S, Dag A** (2004) Sequential introduction of honeybee colonies increases cross-pollination, fruit set and yield of Spadona pear (*Pyrus communis*). *Journal of Horticultural Science and Biotechnology* **79**, 652-658
- Thomson JD, Goodell K** (2001) Pollen removal and deposition by honeybee and bumblebee visitors to apple and almond flowers. *Journal of Applied Ecology* **38**, 1032-1044
- Vansell GH** (1946) Bees and pear pollination. *Proceedings of the Oregon State Society for Horticultural Science* **37**, 51-53

- Vicens N, Bosch J** (2000) Pollinating efficacy of *Osmia cornuta* and *Apis mellifera* (Hymenoptera: Megachilidae, Apidae) on 'Red Delicious' apple. *Environmental Entomology* **29**, 235-240
- Vithanage HINV** (1990) The role of European honeybee (*Apis mellifera* L.) in avocado pollination. *Journal of Horticultural Science* **65**, 81-86
- Webster AD** (2002) Factors influencing the flowering, fruit set and fruit growth of European pears. *Acta Horticulturae* **596**, 699-709
- Westwood NM** (1993) *Temperate Zone Pomology: Physiology and Culture* (3<sup>rd</sup> Edn) Timber Press, Portland, Oregon
- Williams IH** (1994) The dependence of crop production within the European Union on pollination by honeybees. *Agricultural Zoology Review* **6**, 229-257
- Williams RR** (1966) Pollination studies in fruit trees: III. The effective pollination period for some apple and pear varieties. *Annual Report of Long Ashton Agriculture & Horticulture Research Station* **1965**, 136-138
- Williams RR, Smith BD** (1967) Pollination studies in fruit trees. VII. Observation on factors influencing the effective distance of pollinator trees in 1966. *Reports of Agricultural and Horticultural Research Station, University of Bristol* **1966**, 126-134
- Willmer PG, Bataw AAM, Hughes JP** (1994) The superiority of bumblebees to honeybees as pollinators: insect visits to raspberry flowers. *Ecological Entomology* **19**, 271-284
- Zisovich AH, Stern RA, Shafir S, Goldway M** (2004a) Identification of seven *S*-alleles from the European pear (*Pyrus communis*) and the determination of compatibility among cultivars. *Journal of Horticultural Science and Biotechnology* **79**, 101-106
- Zisovich AH, Stern RA, Sapir G, Shafir S, Goldway M** (2004b) The RHV region of *S*-RNase in the European pear (*Pyrus communis*) is not required for the determination of specific pollen rejection. *Sexual Plant Reproduction* **17**, 151-156
- Zisovich AH** (2003) Study on pollination and fertilization factors influencing yield in the Israeli pear orchard. MSc Thesis, submitted to the Faculty of Agricultural, Food and Environmental Sciences, The Hebrew University of Jerusalem, Rehovot, 82 pp
- Zisovich AH, Stern RA, Shafir S, Goldway M** (2005) Fertilization efficiency of semi- and fully-compatible European pear (*Pyrus communis*) cultivars. *Journal of Horticultural Science and Biotechnology* **80**, 143-146
- Zohary M** (1962) *Plant Life of Palestine*, The Ronald Press, NY, USA
- Zur A, Gur A** (2000) *Apple Cultivation*, Israel Fruit Growers Association Publishing, Tel Aviv, Israel (in Hebrew)