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# INSECT POLLINATION OF APPLE ORCHARDS

**THESES OF DOCTORAL (PhD) DISSERTATION**

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# I. INTRODUCTION

The research work concerning the pollination of apple had begun at the beginning of the last century, when the intensification of fruit growing made the growers reduce the number of cultivars and choose only a few, but the most popular cultivars for planting. However, concentration on a few commercial cultivars led to inadequate fruit set and yield. The background of fertilisation problems was studied by several researchers, so the necessity of cross-pollination on apple had also been revealed in the early years of the last century, whereas the importance of the most favourable and appropriate arrangement of cultivars had only been recognised some decades later (SOLTÉSZ, 1996). The questions related to the insect pollination of apple had been started to investigate more intensively even later, in the early years of the fifties (BENEDEK et al., 1974; McGREGOR, 1976; FREE, 1993).

In spite of the fact that the Hungarian agriculture had gone through serious economic depression after the change of regime, apple will remain the most important species of the Hungarian fruit growing in the future (PAPP, 1996).

Based on the results of research it is well known that the pollen and nectar production of apple cultivars determine their attractiveness to honeybees and intensity of bee visits on their flowers. There is a great amount of evidence, which clearly shows that definite differences exist between different apple cultivars in their nectar and pollen production as well (FREE, 1993; BENEDEK, 1996).

There were few indications in the literature that the differences between cultivars concerning their attractiveness to bees, namely their flower characteristics, their nectar and pollen production how could influence the efficiency of flower visiting insects, especially of honeybees. For this reason intensive research was carried out by several authors and it has been proved that not only the differences between the flower structure of cultivars, but the differences between their pollen and nectar production had a definite influence on the honeybee foraging behaviour and on their pollinating efficiency (BENEDEK et al. 1974; DeGRANDI-HOFFMAN et al., 1985; DAVARY-NEJAD et al., 1993; BENEDEK & NYÉKI, 1996/a).

Triploid apple cultivars produce much more pollen per anther than diploid ones, although the pollen grains of triploid cultivars have much lower germination rate (LARSEN & TUNG, 1950). The plenty pollen production can be more important at those cultivars that produce very low amount of nectar. The amount of nectar secreted and its sugar concentration is more changeable than pollen production of the same cultivars, because the nectar secretion is much more influenced by the environmental and weather conditions (RYLE, 1954; FREE, 1993; BENEDEK & NYÉKI, 1996/a). It has been proved that the flowers with all the petals fallen secrete the most concentrated nectar and they are eagerly visited by bees. Consequently, the usage of post-flowering insecticides can increase bee mortality (BENEDEK et al., 1974; WILLIAMS & BRAIN, 1985; COURANT, 1994).

Although the cultivars, whose flowers produce nectar with relatively high sugar concentration, are the most attractive to the bees, the results indicate that the intensity of bee visits first of all depends on the amount of nectar secretion (BENEDEK & NYÉKI, 1996/a).

In planning the most satisfactory combination of compatible cultivars the growers should choose such cultivars whose flowers produce same amount of nectar with approximately same sugar concentration to ensure a well-balanced attractiveness to honeybees (SOLTÉSZ & SZABÓ, 1998).

*Further studies are needed to investigate the daily nectar secretion of different cultivars as well as their attractiveness to pollinating insects, which can help the growers to choose the most appropriate cultivars for planting and to estimate the number of honeybee colonies necessary to pollinate a given apple orchard.*

The *Apoidea* dominates the insect pollinator assemblages on apple and on the overwhelming majority of temperate-zone fruit species. The apple flowers are visited by most groups of apoids that fly in the flowering period of apple. Different wild *Apoidea* species can be valuable and effective pollinators (*Andrena*, *Anthophora*, *Bombus*, *Osmia* spp.), provided that the density of their populations is adequate to ensure good pollination, fruit set and yield (FREE, 1966; BENEDEK et al., 1974).

Honeybees form the highest percentage of flower visiting insect population (70-90%) (FREE, 1966; BENEDEK et al., 1972). The wild bees visiting the apple flowers collect primarily pollen to feed their brood, however, the foraging behaviour of honeybees can be different, they can be pollen gatherers, nectar gatherers and there are honeybees, which collect both pollen and nectar at the same time.

The foraging behaviour of bees determines their efficiency as pollinators. In this point of view, the pure pollen gatherers and bees with mixed behaviour are the most effective pollinators, while pure nectar gatherers do not play significant role in pollination (BENEDEK, 1997). There are certain apple cultivars, whose flowers are characterised by relatively upright stamens round the nectaries, resulting in 'basal gaps' between them, which enable the bees to approach the nectaries from the side without touching the anthers and stigmas. This phenomenon can increase the proportion of the so-called side worker nectar gatherers that are known to be ineffective pollinators (FREE & SPENCER-

BOOTH, 1964; ROBINSON & FELL, 1981; KUHN & AMBROSE, 1982; DeGRANDI-HOFFMAN et al., 1985; BENEDEK & NYÉKI, 1994).
According to BENEDEK & NYÉKI (1996/a), the ratio of pure pollen gatherer honeybees ranges between 40-60%, but the proportion of bees with mixed behaviour (collecting both nectar and pollen) and of pure nectar gatherers can be fairly changeable.
*Further research is needed to investigate the proportion of the different behaviour types of honeybees in the different time of the day at the different cultivars. Further studies are also needed on the effect of nectar secretion and its sugar concentration of the different apple cultivars on the behaviour of bees, especially on the ineffective pollinators, namely, the sideworker nectar gatherers.*

Besides the factors mentioned above, the presence of competitor plants, their simultaneous blooming periods with apple can influence the population and the density of flower visiting insects, consequently the number of honeybee colonies required for pollination in a given apple orchard (BENEDEK et al., 1974; FREE, 1993).
*Therefore, further studies are necessary to evaluate the effect of competitor plant species on the pollinating insects in the different time of the day.*

Every sixth apple flower – approximately 5-10% of the flowers visited by pollinating insects, set fruit (FREE, 1966), although it can be enough to get an adequate fruit set and yield, provided that the insect pollination is ensured.
It has been proved experimentally, that the longer the effective period of bee pollination the higher the fruit set and yield as well as the content of well-developed seeds of fruits (BENEDEK et al., 1974; BENEDEK, 1996; BENEDEK et al., 2000), although the limitation of insect pollination does not have an influence on colouration and quality of fruits (BENEDEK & NYÉKI, 1996/b).
*Further research is needed to describe the relationship between the flower visiting behaviour of honeybees and the fruit set of apple numerically, and to evaluate the effect of limitation of insect pollination on the fruit set and yield of different apple cultivars in the morning and in the afternoon as well.*

We have to ensure not only the suitable pollinizer cultivars, but also the adequate number of honeybee colonies necessary to pollinate an apple orchard and to obtain an economic fruit yield.
The honeybee colonies required in an orchard depend on many factors that vary in different orchards and places (the arrangement and placement of pollinizer cultivars, planting patterns, the presence of pollinating wild bee and feral bee communities, the intensity of bee visits and of course, environmental and weather conditions).
Keeping all these facts in view, one strong honeybee colony per hectare is adequate to pollinate young apple orchards, while three colonies per hectare are necessary in mature orchards. Even more, approximately 3-6 colonies/ha are

recommended in the high-density apple orchards (BENEDEK et al., 1990; BENEDEK, 1997).

To estimate the number of honeybee colonies needed in a given apple orchard, growers should observe the number of bee visits on their fruit trees. If the number of flower visiting bees (on branches bearing 50 flowers observed for 10 minute's period) is about 3-6 on average, it can be adequate to obtain a good fruit set and yield (BENEDEK et al., 1989), but, if the mean number of bee visits is fewer than 2, we have to move further honeybee colonies in the orchards at short notice (PALMER-JONES & CLINCH, 1968).

Both the growers and beekeepers should be made have interested in the controlled bee pollination of fruit orchards, therefore advisory network should be established. There are user-friendly computer simulating methods available to the growers, which can reliably predict the apple yield (FREE, 1993; BENEDEK, 2002). One of these methods, the PC-REDAPOL has also been under development nowadays (DeGRANDI-HOFFMAN, 1995).

There are numerous topics concerning the pollination of apple that greatly need intensive research in the future. Therefore more attention should be paid on analysing the effect of the variety features on the insect pollination, especially in the new type, high density orchards, where the density of insect pollinators, honeybee, wild bee and feral bee populations are much less abundant, than in the traditional, small fruit orchards (FREE, 1993).

To ensure even fruit set with high quality in the large, high-density orchards, whose investment and cultivation costs and are very high, consequently the production should be profitable each year of their short productive life and the conditions of insect pollination should be a stable element of their management system as well.

Based on the above discussion, the aim of our research is to analyse and describe the relationship between the insect pollination and the fruit set of apple numerically, which can help *to estimate the number of honeybee colonies necessary for pollinating a given apple orchard with higher certainty.*

**1. Evaluation of the daily nectar secretion of apple cultivars and their attractiveness to insect pollinators**

**2. Foraging behaviour of honeybees and the intensity of bee visits in the morning and in the afternoon at the examined cultivars**

**3. The effect of nectar production and its sugar concentration of the examined cultivars on the foraging behaviour of bees**

**4. The effect of the foraging behaviour of honeybees on the fruit set, yield and the viable seed content of fruits**

**5. The effect of limitation of insect pollination on the fruit set and yield**

**6. Evaluation of the effect of simultaneously flowering competitor plants with apple on bees**

# II. MATERIAL AND METHODS

## 1. Places and periods of observations

Our measurements were made at two sites in Hungary between 2001 and 2003, in a small experimental orchard with 7-12 year old trees of 18 cultivars in Mosonmagyaróvár (Horticultural Department of the University of West Hungary, Faculty of Agricultural and Food Sciences) and in a commercial plantation where small blocks were available with 10-12 year old trees of 6 cultivars at Feketeerdő.

## 2. Material of observations

Our experiments were made at 18 cultivars.

Mosonmagyaróvár: Akane, Arlet, Braeburn, Early Gold, Florina, Freedom, Gala Must, Gloster, Golden B, Golden Spur, Granny Smith, Idared, Jonagold, Jonagold Wilmuta, Jonathan M-41, Naményi Jonathan, Ozark Gold and Red Elstar.

Feketeerdő: Braeburn, Gloster, Golden B, Idared, Jonagold Wilmuta and Red Elstar.

## 3. Methods of observations

### 3.1. Measurement of nectar production

One branch was chosen at the northern and the southern part of two trees per cultivar and caged with parchment bags to measure nectar production. To measure nectar secretion in the morning and in the afternoon the branches selected were caged from 8-12 a.m. and from 12-16 p.m., respectively. The nectar secretion of 5 flowers per branch was sampled by using the capillary method developed by CRUDEN & HERMANN (1979, 1983) after removing the parchment bags. Amount of nectar was measured by digital scales, its sugar concentration was determined by the Abbe-refractometer in laboratory and the sugar content was calculated afterwards.

## 3.2. Bee pollination of apple

The observations concerning the bee pollination of apple were made by using the method of BENEDEK (1974).
Three strong honeybee colonies per hectare were moved to both orchards just before the flowering had begun.

### 3.2.1. Intensity of bee visitation and the foraging behaviour of honeybees on apple flowers

We made parallel observations at each cultivar examined whose intensity of flowering was approximately similar and their anthesis (anther dehiscence) had begun.
One branch bearing 50 flowers was chosen at the northern and the southern part of two trees per cultivar and bee visitation was observed for tree days (on days with sunny weather being favourable to bee flight). Each branch was observed for 20 minute periods in the morning (8-12 a.m.) and in the afternoon (12-16 p.m.) and the number of bees visiting as well as the number of flowers visited by bees were counted.
Foraging behaviour of bees was also observed at each cultivar. Four kinds of behaviour classes were used related to food gathering behaviour of honeybees: pollen gatherers, nectar gatherers, bees with mixed behaviour (collecting both nectar and pollen) and side worker nectar gatherers. The fifth group was recruited from flower-visiting insects, other than honeybees, their numbers was also registered regardless of their food gathering behaviour.
The number of flowers visited by the whole pollinating insect population was counted as the relative bee visitation (percent). The effective bee visitation calculated excluding the number of flowers visited by the ineffective pollinators (side worker honeybees and the pollinating insects of allotrophic, hemitrophic and harmful distrophic groups).

### 3.2.2. Relationship between the nectar production of apple cultivars and the number of flower visiting insects as well as the foraging behaviour of bees

The effect of nectar production, sugar concentration and sugar content of different cultivars on the foraging behaviour of honeybees was evaluated statistically (at P=5% level).

### 3.2.3. Relationship between the intensity of bee visitation as well as the flower visiting behaviour of honeybees and the fruit set, and the seed content of fruits

Two trees were chosen per cultivar and the fruit set, yield and seed content of fruits were measured on the same branches where the intensity of bee visitation and the foraging behaviour of honeybees was observed.

Treatments were as follows:

**Pollination in the morning:** we left uncovered the previously marked branches and caged them with parchment bags from 12 to 18 o'clock.

**Pollination in the afternoon:** we caged the branches from 6 to 12 o'clock and the parchment bags were removed afterwards from 12 o'clock.

Later fruit set and the number of viable seeds per fruit were measured on the branches. The effect of bee visitation and the foraging behaviour of honeybees on the fruit set and on the seed-content of fruits was evaluated statistically (at P=5% level).

### 3.2.4. The effect of limitation of insect pollination on the fruit set and yield of apple

The effect of flower visiting insects on the fruit yield was studied by limitation of their activity.
To evaluate the fruit yield in the morning and in the afternoon the insect pollination was limited during the certain stages of blooming period or its whole duration. The results of these treatments were compared to the fruit yield on the trees with free pollination.

Treatments were as follows:

**1. No caging** = free pollination.

**2. Caging for the first half of the flowering period** = partial limitation of insect pollination.

**3. Caging for the second half of the flowering period** = partial limitation of insect pollination.

**4. Caging for the two-thirds of the flowering period** = partial limitation of insect pollination.

**5. Caging with parchment bags during the whole flowering period** = total limitation of insect pollination.

The parchment bags were removed after petal fall. Fruit set and yield were measured on branches later, and the mass of fruits as well as the number of viable seeds per apple were also registered.

### 3.2.5. Intensity of bee visitation on the competitor plants

Intensity of bee visits was also observed at competitor plants such as other fruit trees flowering simultaneously, and some flowering weeds.

Three trees were selected at apple trees and at other flowering fruit trees such as cherry, plum, pear etc. and branches bearing 50 flowers were chosen for counting the number of flower visiting insects. Intensity of bee visitation of flowering weeds was also measured by observing the bee flight on them three times in 1 $m^2$ sample squares.

### 3.2.6. Statistical evaluation of the results of observations

The data of our experiments were evaluated by using the Excel 2000 for Windows.

# III. RESULTS

## 1. Evaluation of the daily nectar secretion of apple cultivars and their attractiveness to insect pollinators

Based on our results concerning the nectar production of the examined cultivars, we have corroborated the earlier findings that the amount of nectar secretion of apple flowers, the sugar concentration and sugar content of apple nectar varies between different cultivars and years concerned (FREE, 1993; DAVARY-NEJAD, 1993; BENEDEK és NYÉKI, 1997).

According to our data, the different periods of the day also have an influence on the nectar production – depending on the cultivars - especially the amount of nectar and nectar sugar produced, although the effect of this factor was not significant at both experimental sites.

We found that there were smaller or larger differences between the nectar production of examined cultivars concerning the different periods of the day.

Large differences can be expected between the intensity of bee visitation of the pollinizer and the main cultivars, when great differences exist between their daily nectar production, therefore this fact has to be taken into consideration in orchard planning and estimating the number of honeybee colonies required as well.

*We found that the amount of nectar produced in apple flowers had a positive effect on the number of pollinating insects, that proved to be significant from the statistical point of view (r = 0,42-0,54).*

*The higher the nectar production of a given cultivar the higher intensity of bee visitation can be observed on its flowers* (’Gloster’, ’Jonagold’, ’Jonagold Wilmuta’ and ’Jonathan M 41’). Our results corroborate the earlier statements of BENEDEK és NYÉKI (1996/a). The relationship between the amount of nectar production and the intensity of bee visitation proved to be stronger without taking the ratio of side worker nectar gatherers into account (r = 0,6-0,7). The effect of the amount of nectar secretion on the intensity of bee visitation was higher in the morning than in the afternoon.

Those cultivars that produced nectar with high sugar concentration were more preferred by bees than cultivars whose flowers secreted lower amount of nectar with the same sugar concentration (e.g. ’Florina’, ’Granny Smith’).

*We found that the number of flower visiting insects was less influenced by the sugar concentration of nectar than the amount of nectar produced in the apple* *flowers. Moreover, the sugar concentration of nectar had no influence on the intensity of bee visitation in the afternoon in certain cases (r = -0,07- -0,7). However, relatively higher intensity of bee visitation could be observed at cultivars characterized by abundant nectar production if the sugar concentration of nectar increased in their flowers.*

*There were greater differences between in the attractiveness of the examined cultivars in the morning than in the afternoon.* In the morning twice as many bees visited the cultivars that were most preferred by bees as the flowers of those that were less preferred by them.

It is to be noted that the differences between the attractiveness of the examined cultivars were smaller in the afternoon in spite of the fact that there were also large differences between their nectar production and sugar concretion of nectar both in the morning and in the afternoon.

In the afternoon the bee visitation of the less attractive cultivars was also relatively intense ('Akane, 'Arlet' and 'Early Gold'), compared to the cultivars that were most intensely visited by bees (e. g. 'Braeburn', 'Gloster', 'Jonagold' cvs, 'Jonathan M 41', 'Red Elstar')

## 2. Foraging behaviour of honeybees and the intensity of bee visits in the morning and in the afternoon at the examined cultivars

We found that the half of the flower visiting insect populations was made of pure pollen gatherer honeybees, in the morning and in the afternoon as well - in accordance with the findings of several research workers (BENEDEK et al., 1974; McGREGOR, 1976; HELLMICH & ROTHENBUHLER, 1986; BENEDEK et al., 1989/a; FREE, 1993; BENEDEK & NYÉKI, 1996/a). However, the proportion of pollen gatherers was greater (from 5 to 10%) in the afternoon than in the morning. The ratio of pure nectar gatherers and of bees with mixed behaviour and of side workers was fairly changeable depending on the cultivars examined and on the different periods of the day. The ratio of bees with mixed behaviour and of pure nectar gatherers ranged from 20 to 30% and of nectar gatherers approaching nectaries from the side was from 0 to 20%, respectively (*Figure 1 - Mosonmagyaróvár*). The proportion of side worker bees was higher in the afternoon than in the morning, especially in days with favourable weather.

*The side worker nectar gatherers appeared at each examined cultivar*, but this behaviour was rather frequent at the following varieties: 'Arlet', 'Gala Must', 'Gloster','Golden B', 'Golden Spur', 'Granny Smith' and 'Red Elstar'. (Their ratio ranged from 15 to 22% of the pollinating insect population) (*Figure 1 - Mosonmagyaróvár*).

The cultivars with abundant nectar production were visited the most intensively by bees ('Gloster', 'Jonagold', 'Jonagold Wilmuta' and 'Jonathan') both in the morning and in the afternoon (24-25 pollinating insects per 100 flowers in 20

minute periods). The triploid 'Jonagold' group is known to produce pollen with decreased viability, therefore those cultivars that belong to this group, are not suitable pollinizers in orchard planning. In addition to this, the triploid cultivars are also characterized by abundant nectar secretion, consequently much more intense bee visitation can be expected on them compared to the intensity of bee visits at the other cultivars (SOLTÉSZ, 1997).
Besides the cultivars mentioned above, the cultivars that secreted nectar with relatively high sugar concentration were also intensively visited by bees ('Braeburn', 'Gala Must', 'Golden B', 'Golden Spur', 'Ozark Gold' and 'Red Elstar' with approximately 21-24 pollinating insects per 100 flowers in 20 minute periods).
The cultivars characterized by low amount of nectar with changeable sugar concentration were more or less intensively visited by bees ('Early Gold', 'Florina', 'Freedom' and 'Granny Smith'), however, those cultivars whose nectar secretion was lower with relatively low sugar concentration at the same time, were even less preferred by bees both in the morning and in the afternoon ('Akane' and 'Arlet').
The intensity of bee visitation of examined cultivars was found to be more variable in the morning than in the afternoon. In the morning we measured greater differences between the relative bee visitation of inspected cultivars (from 40 to 80%), than in the afternoon (from 70 to 90%).
The comparison relating to the examined cultivars showed that the proportion of the side worker nectar gatherer honeybees as well as the number of flowers visited by them could be largely different at cultivars inspected. We found that the efficiency of pollination could be decreased by 2-10% at those cultivars whose flowers enable bees to obtain nectar from the side.
The ratio of pollinating insects other than honeybees ranged from 1 to 5% on flowers of each cultivar, however, their proportion was also higher in the afternoon (*Figure 1 - Mosonmagyaróvár*). The wild *Apoidea* formed the highest percentage (from 90 to 95%) of this class. Various *Diptera* had also been observed on apple trees, but their population was not considerable at all. The rest of the other pollinating insects was recruited from the distrophic group (harmful beetle species such as *Cetonia aurata*, *Epicometis hirta*) but their population was also not abundant.

### 3. The effect of nectar production and its sugar concentration of the examined cultivars on the foraging behaviour of bees

The amount of nectar secreted and its sugar concentration had a definite influence on the behaviour of honeybees.
*Our results showed that the amount of nectar produced in apple flowers had a positive effect on the number of* ***pure pollen gatherer honeybees*** *and of* ***bees with mixed behaviour*** *both in the morning and in the afternoon, even though*

*these behaviour groups collect pollen solely or primarily from the apple flowers.*
*Both the pure pollen gatherers and bees with mixed behaviour preferred the cultivars with abundant nectar secretion to the ones with relatively low nectar production* (*Figure 2-3 - Mosonmagyaróvár*).
The relationship between the amount of nectar produced and the number of pure pollen gatherer honeybees proved to be strong (r = 0,6-0,7), however, the effect of nectar production was even stronger on bees with mixed behaviour (r = 0,8-0,9), because this group collected mainly nectar from the apple flowers.
The cultivars whose flowers secreted nectar with higher sugar concentration, were less intensively visited by pure pollen gatherer honeybees and bees with mixed behaviour as well.
*The cultivars that produced nectar with the highest sugar concentration, were the most preferred by* ***pure nectar gatherer honeybees*** *in contrast to the two behaviour groups mentioned above* ('Braeburn', 'Gala Must', 'Golden B', 'Golden Spur', 'Granny Smith' and 'Idared') (*Figure 4 - Mosonmagyaróvár*). The results of our observations corroborate the earlier statements described by BENEDEK and NYÉKI (1996/a). The 'Gloster', 'Jonagold', 'Jonagold Wilmuta' and 'Jonathan M 41' cultivars were much less intensively visited by pure nectar gatherer bees, because of the relatively low sugar concentration of their nectar. The relationship between the sugar concentration of apple nectar and the number of pure nectar gatherer honeybees was highly significant from the statistical point of view (r = 0,78-0,91).
***The side worker nectar gatherer honeybees*** *likewise pure nectar gatherers visited also much more intensively those cultivars that produced nectar with higher sugar concentration. It is stressed that the correlation was fairly changeable depending on the experimental sites* (r = 0,23-0,73) (*Figure 5 – Mosonmagyaróvár*).
According to our results, not only the flower morphology of apple flowers but the sugar concentration of nectar has a positive effect on the number of side worker nectar gatherers, especially in the afternoon, when the sugar concentration is higher because of the sunny, dry weather.
The amount of nectar production and its sugar concentration had a little influence on the number of **pollinating insects other than honeybees (wild bees)**, and in certain cases, only (nectar production: r = 0,4-0,76, sugar concentration: r = -0,26 - -0,73). The reason for it partly that their proportion is very low compared to the whole flower visiting insect population (0,5-5%), partly because the different wild bee species fly in the early part of spring when they need pollen to feed the larvae, accordingly, they collect mainly or solely pollen from the apple flowers.

### 4. The effect of the foraging behaviour of honeybees on the fruit set, yield and the viable seed content of fruits

The pollinating insects belonging to the different behaviour groups influenced the fruit set and the number of viable seeds per apple in a different way.

*The results showed that the greatest percentage of fruit set and the highest number of viable seeds per fruit were measured on branches of those cultivars that were visited the most frequently by* ***pure pollen gatherer bees*** *as well as* ***bees collecting both nectar and pollen*** ('Braeburn', 'Gloster', 'Idared', 'Jonagold', 'Jonagold Wilmuta', 'Jonathan M 41', 'Nagyényi Jonathan', 'Ozark Gold' and 'Red Elstar'). The fruit set of cultivars mentioned above was more than 5-10%, and their fruits had 8-9 viable seeds (*Figure 6-9 – Mosonmagyaróvár*).

The effect of pure pollen gatherers and of bees with mixed behaviour on the fruit set and the number of viable seeds per fruit was highly significant from the statistical point of view (pure pollen gatherers – fruit set: r = 0,75-0,93, viable seeds: r = 0,66-0,91; bees with mixed behaviour – fruit set: r = 0,64-0,72, viable seeds: r = 0,66-0,74).

*Those* ***bees*** *that were* ***sucking nectar only*** *on apple flowers did not proved to be effective pollinators at all. Relationship between their number and fruit set as well as the number of well-developed seeds per fruit was not significant*, because the coefficient of correlation was close to nil, however, all nonsignificant figures were negative (fruit set: r = 0,01 - -0,34, viable seeds: r = 0,03 - -0,27).

The reason for the pure nectar gatherer honeybees failed to transfer pollen effectively might be the fact that they collected nectar from flowers with stigmas not receptive yet or already lost their receptivity. It can also be a possible reason that the anthers did not begin to dehisce at those flowers that the bees collected for nectar, and the pollen grains on the bodies of nectar gatherer bees might had a decreased viability or the stigmas did not receive adequate amount of pollen from body hair of nectar gatherer honeybees.

The negative effect of the side worker nectar gatherer bees on the fruit set and the number of viable seeds of fruits has been mentioned by several research workers in the literature (ROBERTS, 1945; PRESTON, 1949; FREE, 1960/b; FREE & SPENCER-BOOTH, 1964/a; ROBINSON, 1979/a; ROBINSON & FELL, 1981; KUHN & AMBROSE, 1982; DeGRANDI-HOFFMANN et al., 1985; BENEDEK & NYÉKI, 1994; THORP, 2000).

*We also found that the ratio of* ***side worker nectar gatherer honeybees*** *was in a negative correlation with the fruit set and the number of viable seeds of apple cultivars examined both at Mosonmagyaróvár and Feketeerdő* (fruit set: r = -0,52 - -0,65, viable seeds: r = -0,45 - -0,85) (*Figure 10-11 – Mosonmagyaróvár*). Their presence led to higher decrease of fruit set and of the amount of

viable seeds per fruit at Feketeerdő than at Mosonmagyaróvár, especially in the morning.
It is to be noted that the most effective pollinators, namely the pure pollen gatherers and the bees with mixed behaviour visited the apple flowers much less intensively in the morning than in the afternoon, therefore the negative influence of side worker nectar gatherers was more remarkable in the morning, even if their number or proportion was smaller.
*The effect of activity of* ***other pollinating insects*** *was found to be fairly variable according to the time of the day.* In the morning they had no effect on the fruit set as well as on the number of viable seeds of fruits either at Mosonmagyaróvár or at Feketeerdő (fruit set: r = 0,2-0,44, viable seeds: r = 0,33-0,46), but in the afternoon, when their activity was more intense, the correlation was a bit stronger (fruit set: r = 0,54-0,8, viable seeds: r = 0,46-0,49) (*Figure 12-13 – Mosonmagyaróvár*).
According to the statements of several researchers, the populations of the wild pollinating insects are rather changeable and are depending on the growing localities, the environmental conditions and the size of the nesting habitats as well. In addition to this, when apple starts to flower, their populations themselves are not abundant yet to pollinate the apple orchards sufficiently (MENKE, 1951 cit. FREE, 1993; FRILLI et al., 1983; BENEDEK, 1992). However, our results showed that the wild pollinating insects (especially the wild *Apoidea*) can contribute to the adequate pollination of apple orchard, even if their populations are relatively low.

## 5. The effect of limitation of insect pollination on the fruit set and yield

Our results clearly showed that the effective duration of insect pollination definetely affected the fruit set, the average mass of fruits as well as the viable seed content of apples (*Figure 14-19 – Mosonmagyaróvár and Feketeerdő*).
Free pollination produced the highest fruit set, mass of fruits and viable seeds of fruits, but total limitation of insect pollination resulted in no fruit yield at all, the examined partially self-fertile cultivars, namely 'Granny Smith', 'Idared' and 'Jonagold Wilmuta' (SOLTÉSZ, 1996/c; SOLTÉSZ & SZABÓ, 1998) did not set fruit, either.
**Free pollination.** The fruit set of each cultivar inspected was more than 5-10%, the average number of viable seeds per apple was 7-9, even at those cultivars that were less favourable for bees. In the afternoon the free pollination resulted in greater fruit set, furthermore, the average mass of apples was higher and the fruits had more well-developed seeds (morning – fruit set: 4-10%, viable seeds: 6-9 per fruit; afternoon – fruit set: 8-18%, viable seeds: 7-10 per fruit). Our results also showed that the recommended 3 honeybee colonies per hectare

(GULYÁS, 1983; BENEDEK et al., 1989/b; GUPTA et al., 1993) proved to be enough to get an adequate fruit yield at both experimental sites – in the case of free pollination.

**Caging for the first half of the flowering period.** This treatment resulted in large decrease of fruit set (-45- -87%, compared to the mean values of free pollination), of average mass of fruits and the number of viable seeds per apple as well (compared to the mean values of free pollination - average mass of fruits: -2 - -10%; number of developed seed per fruit: -3 - -27%).

**Caging for the second half of the flowering period.** On branches covered during the second half of the flowering period, the number of apples, the average mass of fruits and the number of viable seed per apple was higher than on branches that were covered during the first half of the flowering period. However, this treatment gave lower yield than free pollination (compared to the mean values of free pollination the decrease of fruit set was: -12 - -53%, average mass of fruits: -0,6- -6% and viable seeds per apple: -2 - -15%, respectively).

**Caging for the two-thirds of the flowering** period also resulted in decrease of fruit yield, but the yield differences were not so great and nonsignificant compared to the fruit yield on branches left uncovered during the whole flowering period (compared to the mean values of free pollination the decrease of fruit set was: -5 - -33%, average mass of fruits: -0,3 - -5% and viable seeds per apple: -0,8 - -12%, respectively).

It is to be noted the negative effect of limitation of insect pollination on the fruit set and yield was not so great at the overwhelming majority of the examined cultivars in the morning. The reason for it partly that in the morning the weather conditions usually are not favourable for bee flight, partly the receptivity of stigmas of apple flowers and the pollen presentation is not so high as well as the pollen tube growth is also retarded because of the low temperatues. Accordingly, in the afternoon when the intensity of bee visitation is evidently higher, the limitation of insect pollination can lead to higher decrease of fruit set and yield.

Besides this, in the morning the proportion of side worker bees was higher compared to the whole pollinating insect population at those cultivars whose flowers enable bees to obtain nectar from the side (e.g. ’Arlet’, ’Granny Smith’ and ’Golden B’), consequently, the effectiveness of pollination is always lower in the morning than in the afternoon. In the afternoon when the intensity of bee visitation is higher due to the more favourable weather conditions, the negative effect of side worker nectar gatherers is negligible on the fruit set and, even if their proportion is relatively high yield (e.g. ’Red Elstar’).

## 6. Evaluation of the effect of the simultaneously flowering competitor plants with apple on bees

Some fruit trees and weeds were in flower during the flowering period of apple so they affected the insect visitation of the apple orchards at both experimental sites and each year of our experiments.

Bee activity was especially intense at sweet cherry trees and on dandelion flowering simultaneously. In certain years the attractiveness of plum and rape to bees was higher than that of apple. Sour cherry trees were also frequently visited by bees, however, the attractiveness of pear trees, black currant and red currant bushes was relatively low.

It is to be noted that the examined cultivars were intensively visited by bees on average, therefore the effect of the competitor plants were negligible on the pollinating insects and resulted in decrease of yield at only those cultivars that were the least intensively visited by bees.

To sum up, in accordance with the data available in the literature (BENEDEK, 1997), placement of additional bee colonies in the apple orchards is needed when any of the competitor plants are visited many more bees than apple.

# IV. RECOMMENDATIONS FOR THE THEORETICAL AND PRACTICAL USE OF NEW RESULTS

The results of our research work related to the insect pollination of apple can help to estimate the number of honeybee colonies required for pollination of apple orchards.

1. The effect of the differences between the amount of nectar production of the examined cultivars – especially of the main and pollinizer cultivars – can lead to inadequate pollination as a consequence of the different attractiveness of their flowers to bees that proved to be more disadvantageous in the morning.

2. The side worker nectar gatherer honeybees were observed at each inspected cultivar. In the morning when the intensity of insect visitation was lower than in the afternoon, the proportion of side worker bees was relatively higher compared to the whole pollinating insect population, accordingly, the effectiveness of pollination was proved to be lower in the morning than in the afternoon.

*According to our results, the differences between the nectar production of the main and the pollinizer cultivars as well as the presence of the side worker nectar gatherer honeybees have to be taken into account more carefully in estimating the number of honeybee colonies necessary to pollinate a given apple orchard, especially in the morning and at those cultivars that enable bees to gather nectar from the side.*

*If large differences can be observed between the effective bee visitation of the main and the pollinizer cultivars (in the morning: 30-40%, in the afternoon: 15-20%), further honeybee colonies are needed to place in the apple orchards (1-2 honeybee colonies/ha) in addition to the recommended 3-6 honeybee colonies per hectare.*
*- The honeybees are known to tend to fly and visit the flowers of those trees that there are within in one row and they rarely move to another row. To take this fact into account, the beehives should be placed between the rows of those*

*cultivars that are less preferred by bees, because the honeybees usually visit the flowers of the nearest apple trees to their beehives.*
*- If the intensity of bee visitation at the cultivars is expected to be fairly similar or the main cultivars are a bit more preferred by bees – their effective bee visitation is 10-15 percent higher - the honeybee colonies should be moved in the rows of those cultivars that are suitable pollinizers for most of the main cultivars.*
*- Honeybee colonies should not be placed in the rows of cultivars whose flowers visited much more intensively by bees than the others, because the flower constancy of honeybees can be extremely high on these cultivars, consequently the bees fail to pollinate the other cultivars.*
*- To ensure even pollination throughout an apple orchard with disadvantageous combinations of varieties, the beehives should be moved in the rows of those cultivars that are the least preferred by bees.*

3. We described the relationship between the amount of nectar production and its sugar concentration of the examined apple cultivars and the foraging behaviour of honeybees, numerically.
- The amount of apple nectar secreted had a definite influence on the number of the pure pollen gatherer honeybees and of bees with mixed behaviour (collecting both nectar and pollen) in the morning and in the afternoon as well.
- The flower visiting intensity of the pure nectar gatherers and the side worker nectar gatherer honeybees was definitely higher at those cultivars whose flowers secreted nectar with relatively high sugar concentration.
- The effect of the amount of nectar and its sugar concentration on the pollinating insects other than honeybees (wild bees) was not proved to be significant from the statistical point of view.

4. The effect of bee visitation and foraging behaviour of honeybees on the fruit set and on the seed content of fruits was also evaluated statistically.
- The effect of pure pollen gatherers and of bees with mixed behaviour was highly significant from the statistical point of view on the fruit set and the number of viable seeds per fruit.
- Those bees that were collecting nectar only from apple flowers, were not proved to be effective pollinators at all. The ratio of side worker nectar gatherers was negatively correlated with the fruit set and the viable seed content of fruits of the examined apple cultivars.
- The effect of flower visiting intensity by pollinating insects other than honeybees was found to be fairly variable according to the time of the day. In the morning they had no effect on the fruit set as well as on the seed content of fruits either at Mosonmagyaróvár or at Feketeerdő. However, in the afternoon, when their intensity was greater, the correlation was a bit stronger.

*Our results can help to estimate the number of honeybee colonies necessary to pollinate a given apple orchard. Not only the intensity of bee visitation but the foraging behaviour of honeybees should be taken into account in estimating the number of honeybee colonies required.*
*- According to our data, the pure pollen gatherer honeybees and the bees with mixed behaviour proved to be the most effective pollinators, their ratio ranged from 60 to 80%. The decrease of efficiency of pollination was measured at those cultivars that were visited less intensively by pure pollen gatherers and bees with mixed behaviour. The ratio of these behaviour groups was less than 60-65% at the following cultivars:* 'Arlet', 'Gala Must', 'Golden B', 'Golden Spur', 'Granny Smith' *and* 'Red Elstar'*, however, the low proportion of the pure pollen gatherer honeybees and bees collecting both nectar and pollen resulted in high decrease of efficiency of pollination only in the morning when each cultivar was much less intensively visited by bees than in the afternoon.*
*In this case, further honeybee colonies necessary to move in the apple orchards or the beehives should be supplied with pollen traps that collect pollen grains from the bodies of bees, consequently, the bees are forced to collect much more pollen from the apple flowers to feed their larvae and they can pollinate the orchards more effectively.*
*- The effect of pure nectar gatherer honeybees on the fruit set and the viable seed content of fruits was not significant at all, although, the side worker nectar gatherers had a negative effect on the fruit set and yield. The ratio of these behaviour groups ranged from 10 to 45% at the examined cultivars.*
*- The relatively high ratio of side worker nectar gatherer honeybees (15-25%) resulted in the decrease of effectiveness of pollination at those cultivars that encouraged bees to gather nectar from the side (*'Arlet', 'Florina', 'Gala Must', 'Golden B', 'Golden Spur', 'Granny Smith' *and* 'Red Elstar'*) and especially in the morning when the intensity of bee visitation proved to be low (40-50%). The bee colonies should be placed in the rows of those cultivars that are intensively visited by the ineffective pollinator side worker honeybees.*

5. We found that the limitation of insect pollination led to higher decrease of fruit set and yield in the afternoon. The peak period of pollen presentation of apple is in the afternoon, approximately between 12 to 16 o'clock, when the weather conditions are more favourable for bee flight, consequently, the negative effects of limitation of insect pollination resulted in higher decrease of the fruit set, the average mass of fruits as well as the viable seeds per apple than in the morning.

6. Based on our data, the effect of the competitor plants has to be taken into account more carefully in the morning when the apple flowers less intensively visited by bees compared to the some flowering fruit trees and weeds, especially sweet cherry trees and dandelion.

*Our results clearly showed that the effect of limitation of insect pollination resulted in inadequate fruit set and yield that can only be avoided by placing additional honeybee colonies.*

*Bee attractivity of the other flowering fruit trees can be compensate by moving further bee colonies to the orchards and the weeds can be eliminated by mowing them within and around the apple orchards.*

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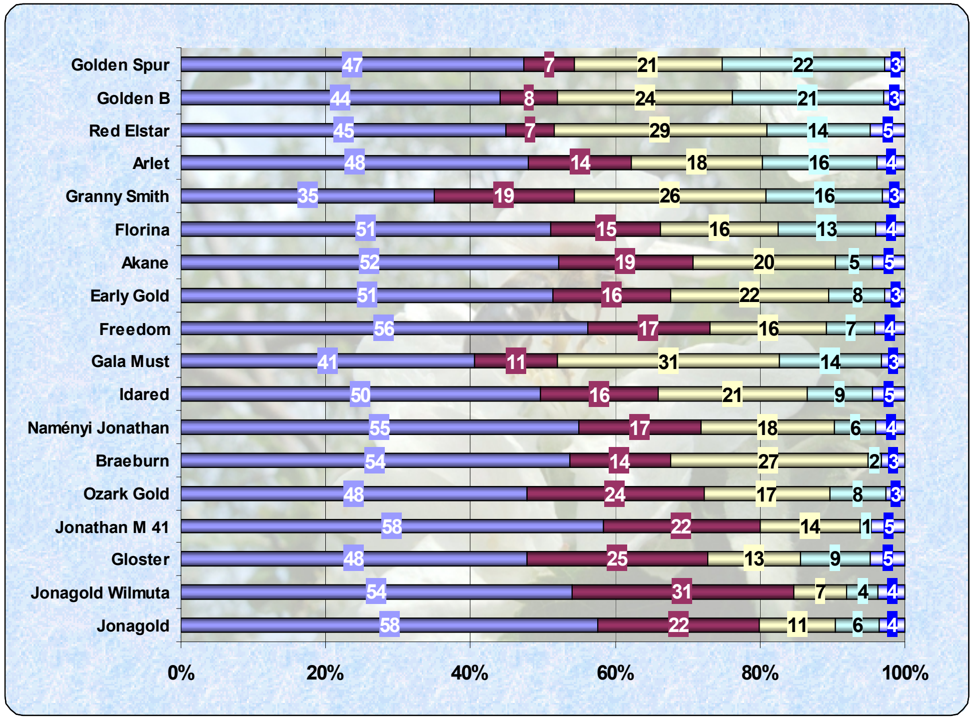
# APPENDIX

***Figure 1*** Foraging behaviour of honeybees and the ratio (%) of pollinators other than honeybees at apple cultivars (Mosonmagyaróvár, 2001-2003)




lilac = pollen gatherer bees, purple = bees with mixed behaviour, yellow = nectar gatherer bees, light blue = side worker nectar gatherer bees, white = pollinators other than honeybees

***Figure 2*** The effect of nectar production on the number of pollen gatherer honeybees visiting the flowers of apple cultivars (Mosonmagyaróvár, 2001-2003)
n = 1296, r = 0,66, significant at P=5% level

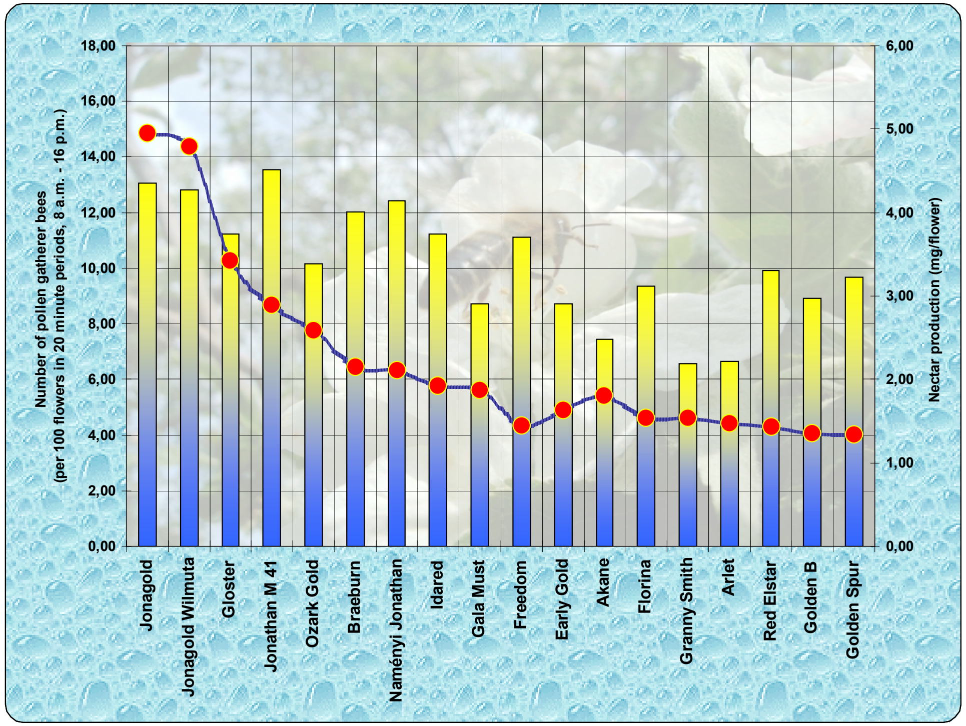


***Figure 3*** The effect of nectar production on the number of honeybees with mixed behaviour visiting the flowers of apple cultivars (Mosonmagyaróvár, 2001-2003)
n = 1296, r = 0,85, significant at P=5% level

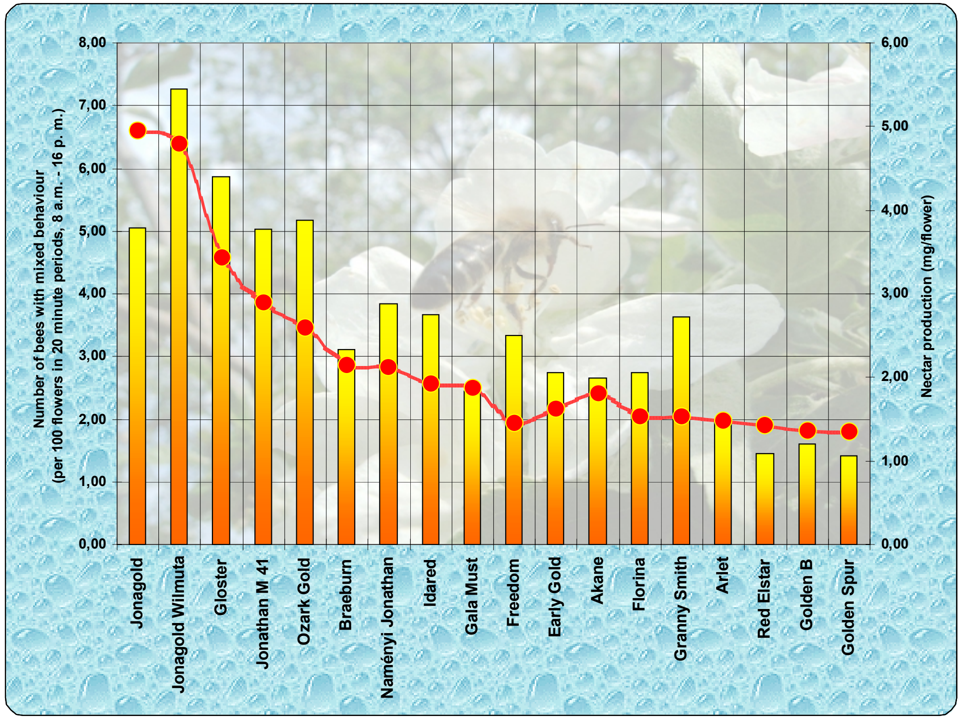


***Figure 4*** The effect of sugar concentration of nectar on the number of nectar gatherer honeybees visiting the flowers of apple cultivars (Mosonmagyaróvár, 2001-2003)
n = 1296, r = 0,78, significant at P=5% level

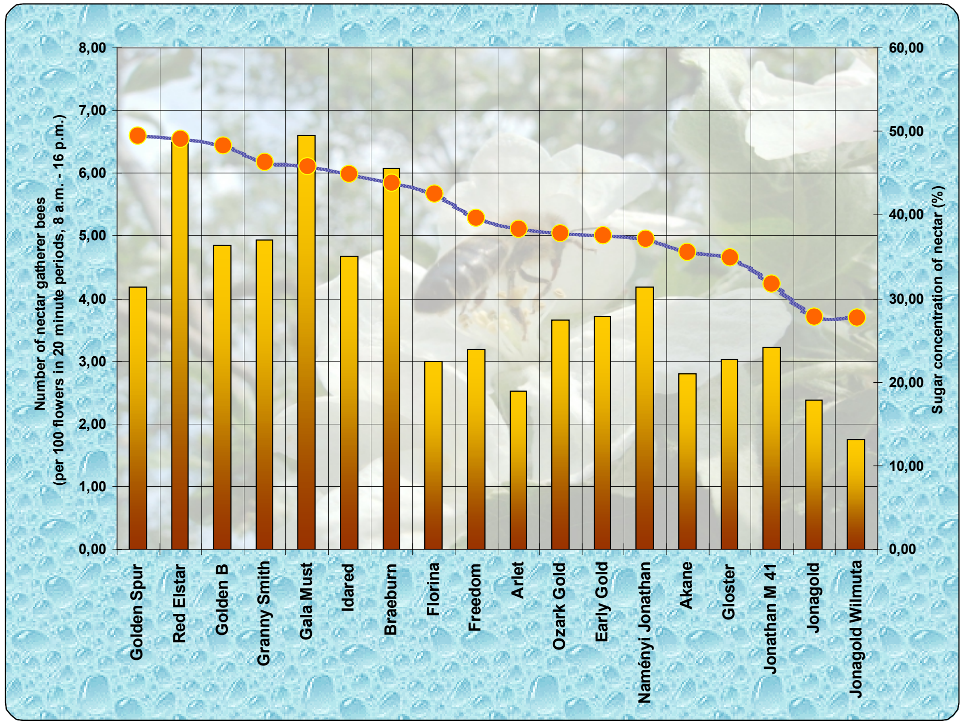


***Figure 5*** The effect of sugar concentration of nectar on the number of side worker nectar gatherer honeybees visiting the flowers of apple cultivars
(Mosonmagyaróvár, 2001-2003) n = 1296, r = 0,73, significant at P=5% level

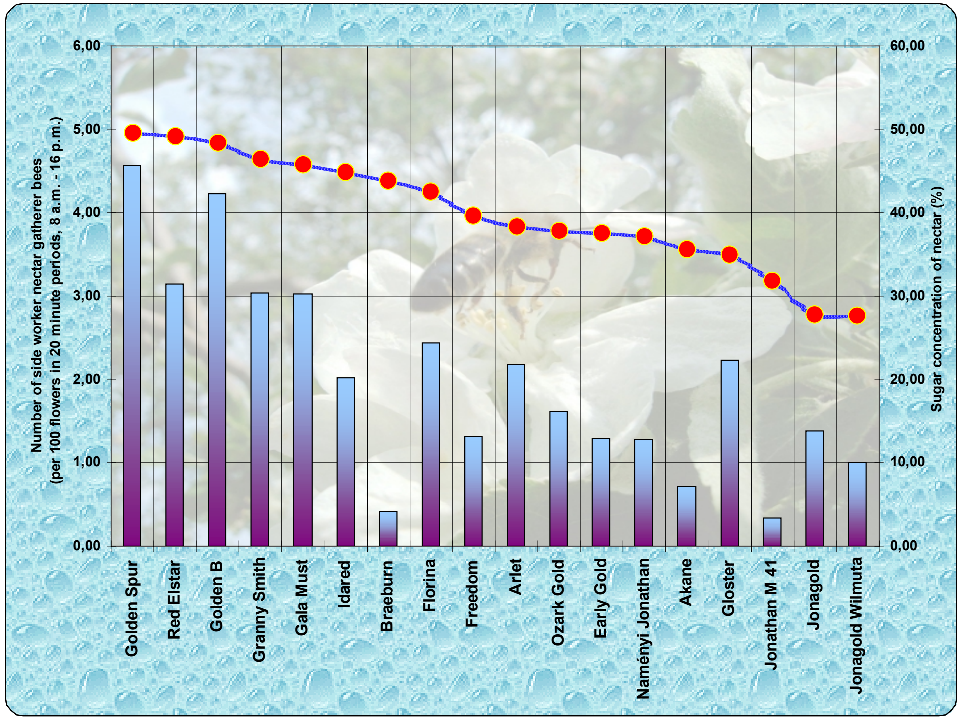


***Figure 6*** The effect of pollen gatherer honeybees on the fruit set of apple (Mosonmagyaróvár, 2001-2003), n = 432, significant at P=5% level

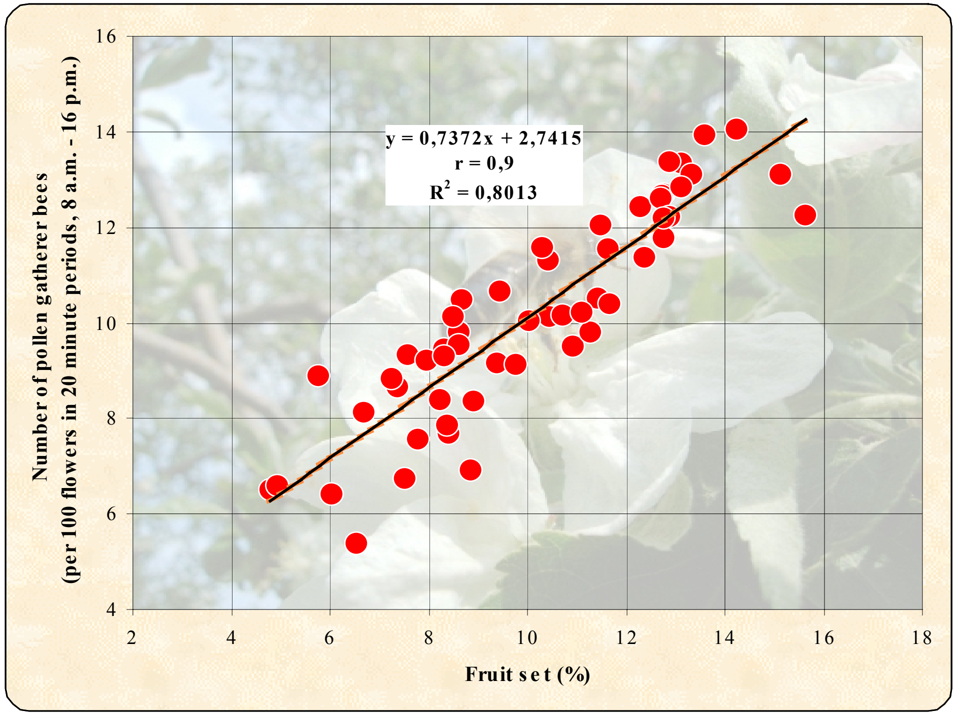


***Figure 7*** The effect of pollen gatherer honeybees on the number of viable seeds per apple (Mosonmagyaróvár, 2001-2003), n = 432, significant at P=5% level

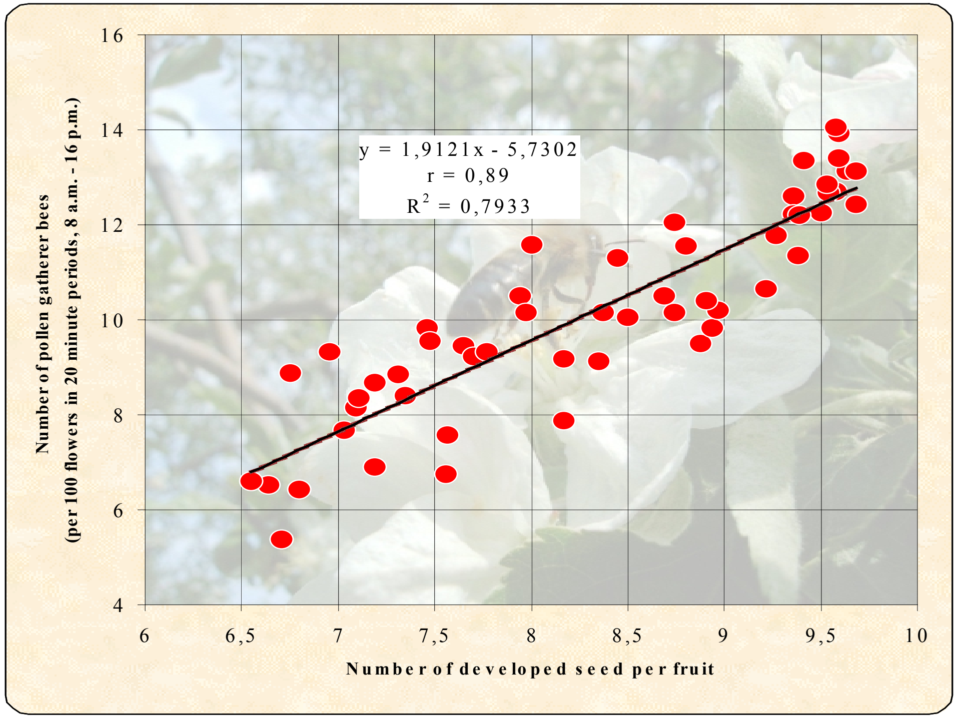


***Figure 8*** The effect of honeybees with mixed behaviour on the fruit set of apple (Mosonmagyaróvár, 2001-2003), n = 432, significant at P=5% level

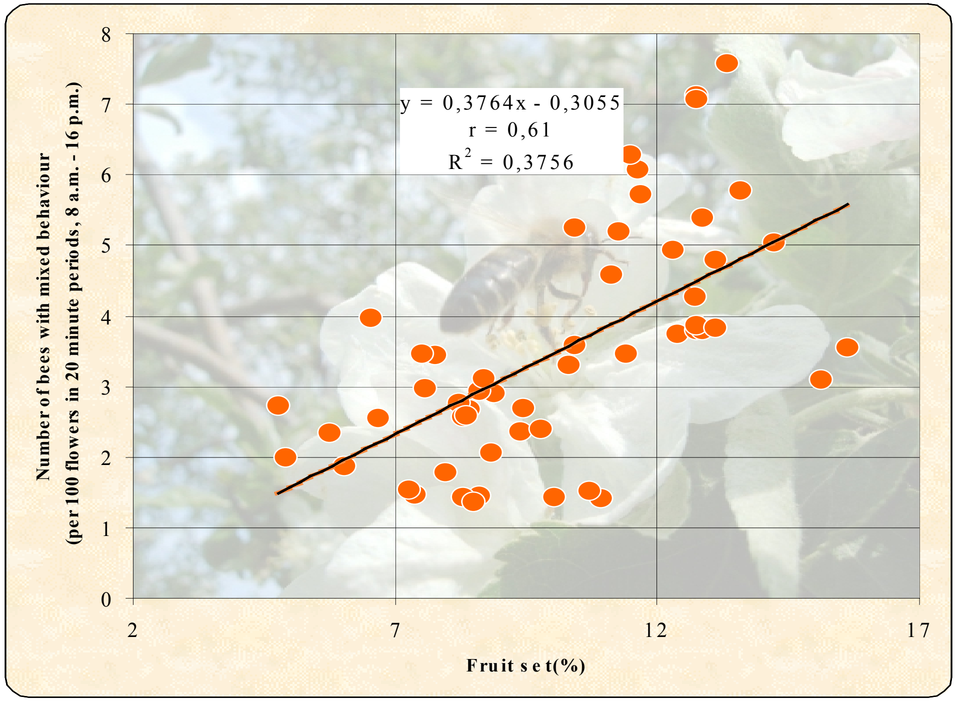


***Figure 9*** The effect of honeybees with mixed behaviour on the number of viable seeds per apple (Mosonmagyaróvár, 2001-2003), n = 432, significant at P=5% level

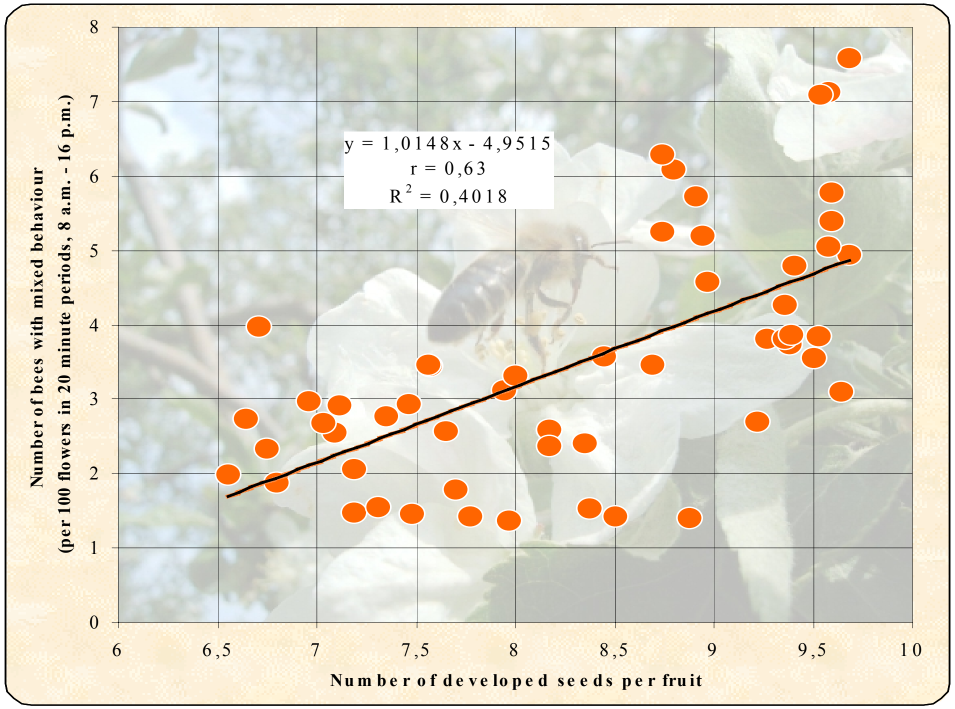


***Figure 10*** The effect of side worker honeybees on the fruit set of apple (Mosonmagyaróvár, 2001-2003), n = 432, significant at P=5% level

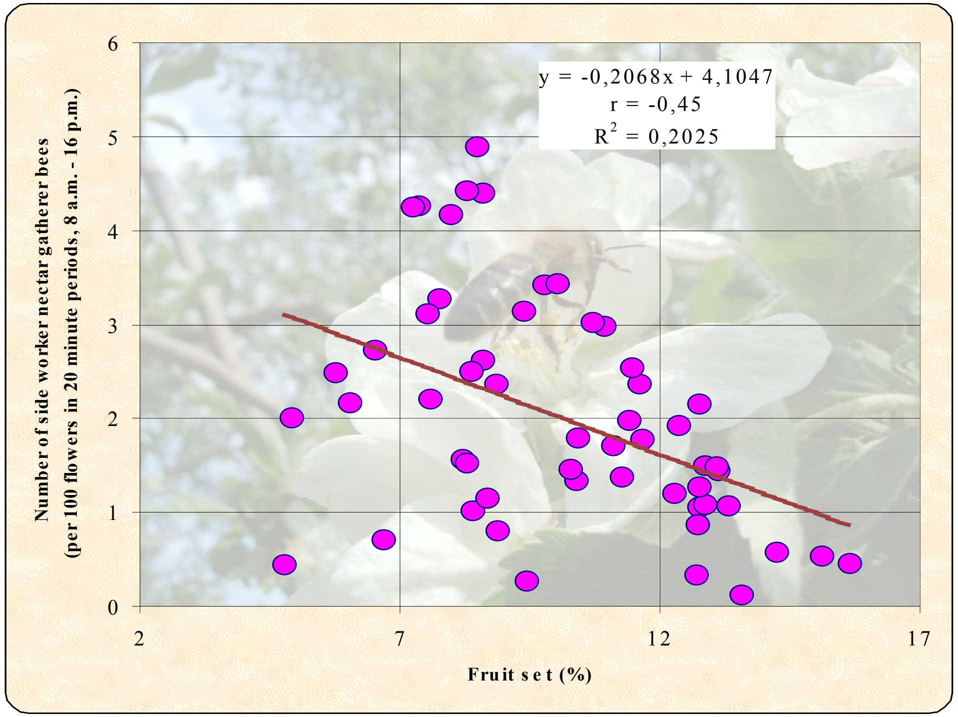


***Figure 11*** The effect of side worker honeybees on the number of viable seeds per apple (Mosonmagyaróvár, 2001-2003), n = 432, significant at P=5% level

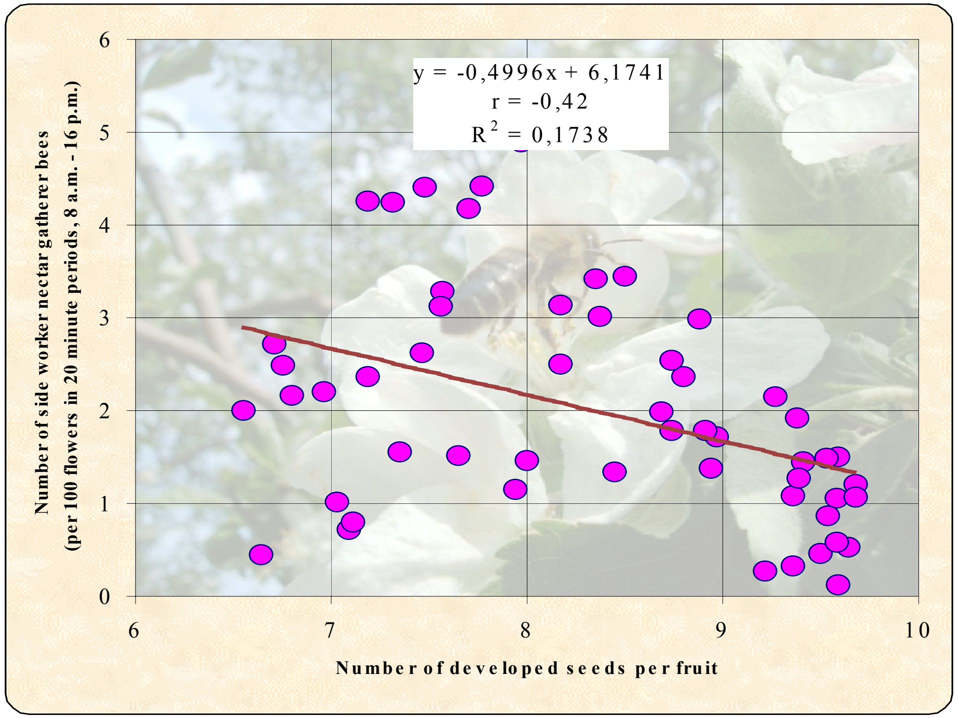


***Figure 12*** The effect of pollinating insects other than honeybees (wild bees) on the fruit set of apple (Mosonmagyaróvár, 2001-2003), n = 432, significant at P=5% level

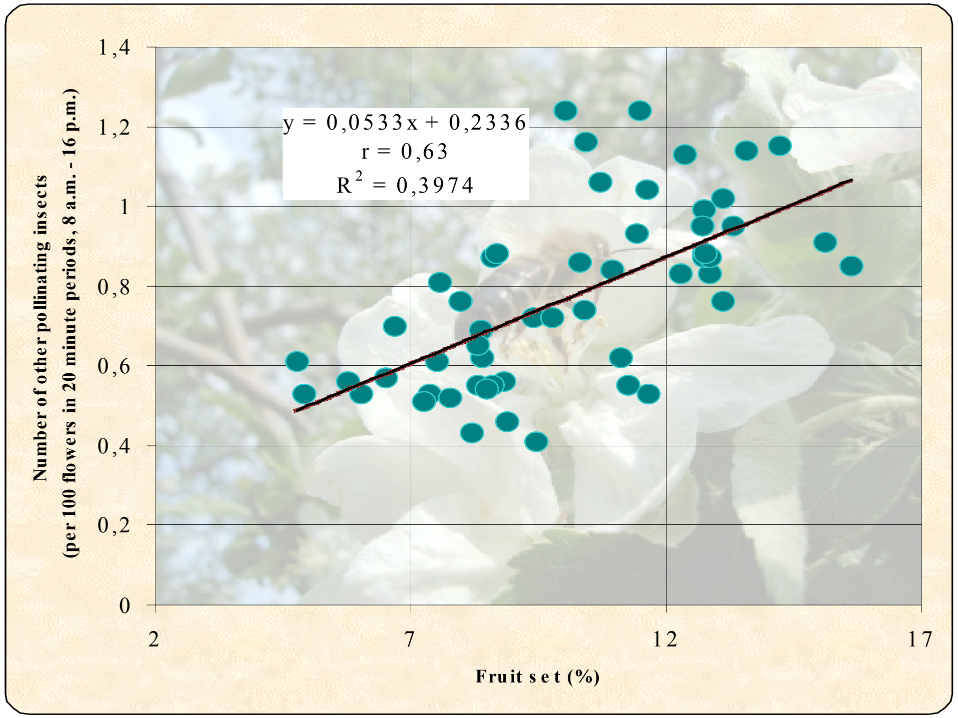


***Figure 13*** The effect of pollinating insects other than honeybees (wild bees) on the number of viable seeds per apple (Mosonmagyaróvár, 2001-2003), n = 432, significant at P=5% level

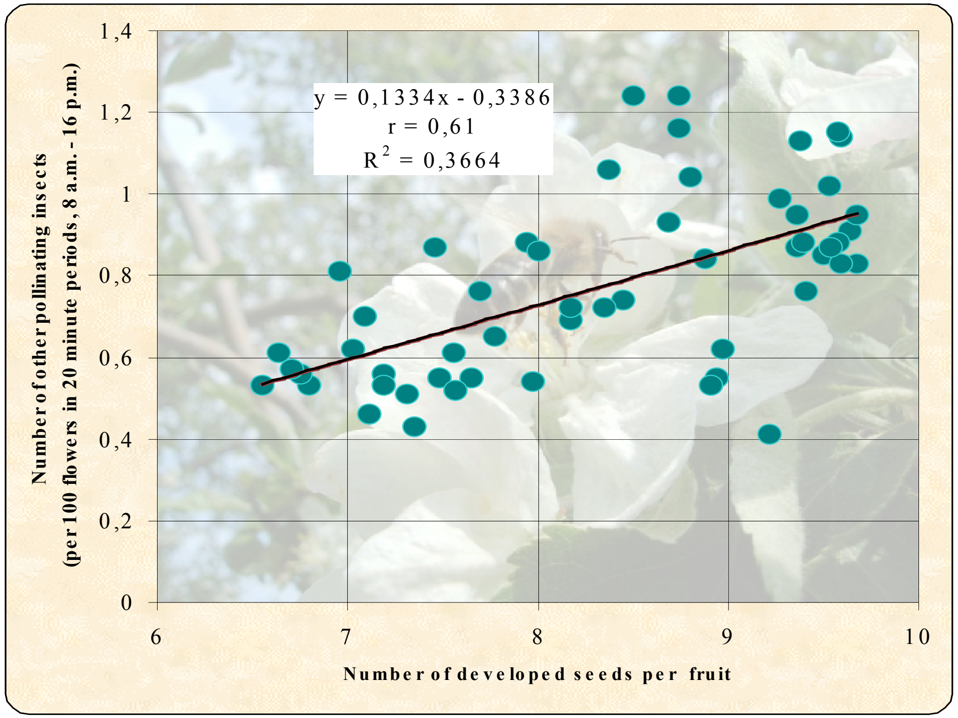
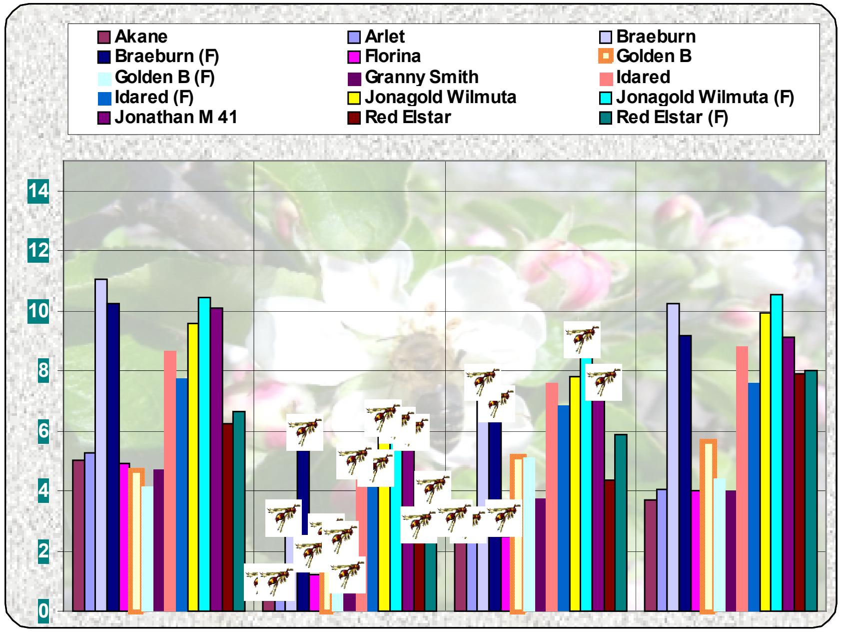


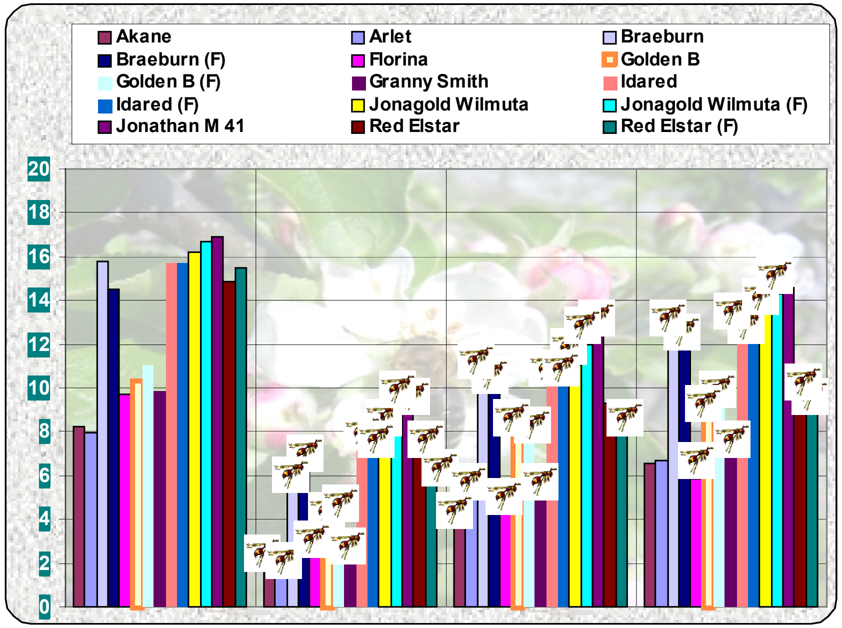
*Figure 14* Fruit set of apple cultivars as affected by the duration of insect pollination in the morning (Mosonmagyaróvár and Feketeerdő (F), 2001-2003)




1. **Free pollination = no caging**
2. **Caging for the first half of the flowering period**
3. **Caging for the second half of the flowering period**
4. **Caging for the two-thirds of the flowering period**

 **There is a statistically significant difference compared to the mean values of 1. (significant at P=5% level)**

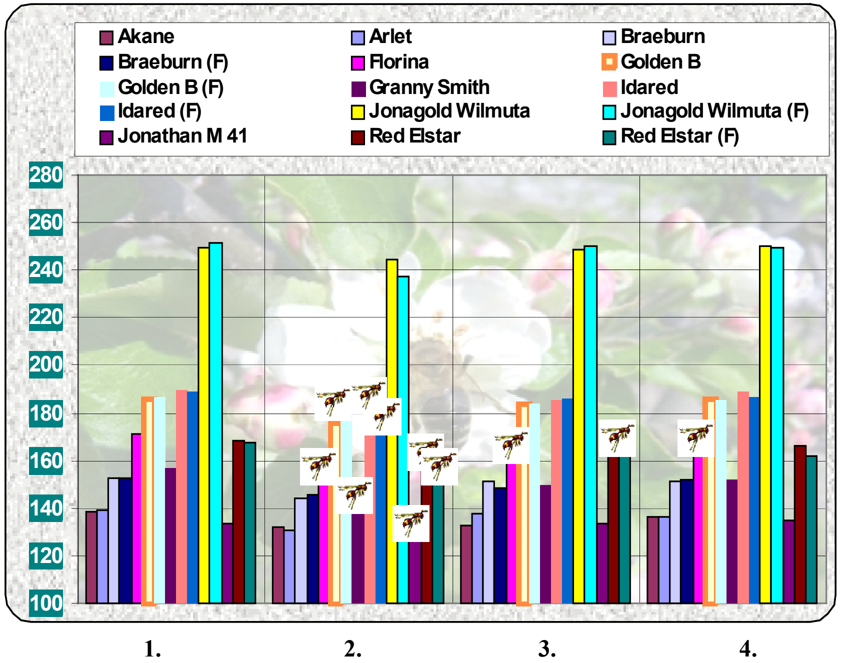
***Figure 15*** Fruit set of apple cultivars as affected by the duration of insect pollination in the afternoon (Mosonmagyaróvár and Feketeerdő (F), 2001-2003)




1. **Free pollination = no caging**
2. **Caging for the first half of the flowering period**
3. **Caging for the second half of the flowering period**
4. **Caging for the two-thirds of the flowering period**

 **There is a statistically significant difference compared to the mean values of 1. (significant at P=5% level)**

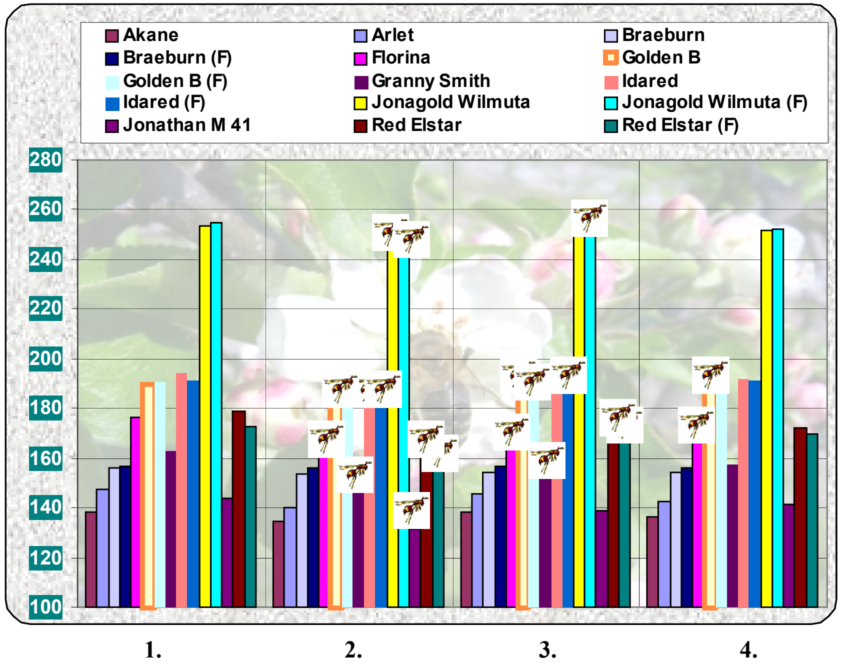
***Figure 16*** Average mass of fruits of apple cultivars as affected by the duration of insect pollination in the morning (Mosonmagyaróvár and Feketeerdő (F), 2001-2003)




1. **Free pollination = no caging**
2. **Caging for the first half of the flowering period**
3. **Caging for the second half of the flowering period**
4. **Caging for the two-thirds of the flowering period**

**There is a statistically significant difference compared to the mean values of 1. (significant at P=5% level)**

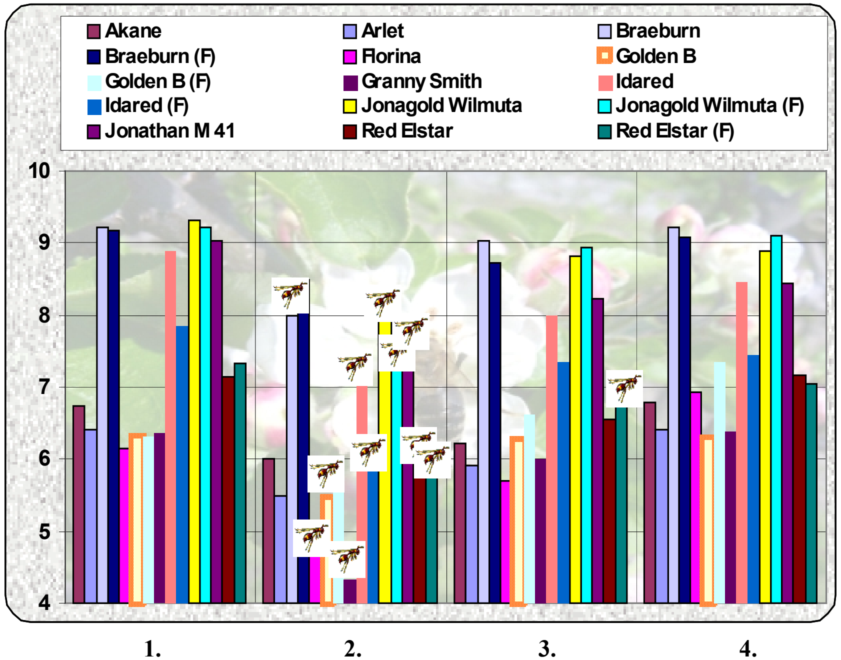
***Figure 17*** Average mass of fruits of apple cultivars as affected by the duration of insect pollination in the afternoon (Mosonmagyaróvár and Feketeerdő (F), 2001-2003)




1. **Free pollination = no caging**
2. **Caging for the first half of the flowering period**
3. **Caging for the second half of the flowering period**
4. **Caging for the two-thirds of the flowering period**

**There is a statistically significant difference compared to the mean values of 1. (significant at P=5% level)**

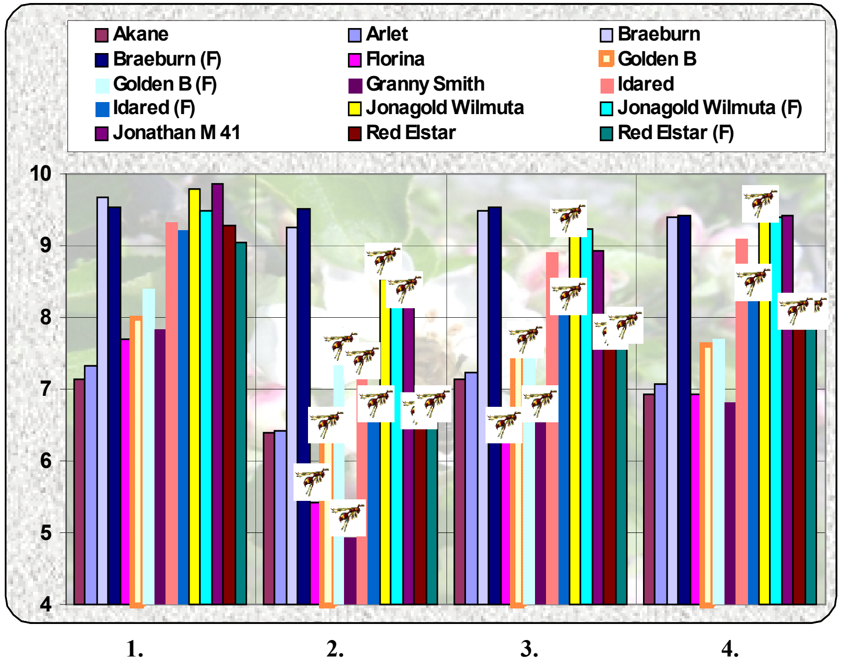
***Figure 18*** Number of viable seeds per fruit of apple cultivars as affected by the duration of insect pollination in the morning
(Mosonmagyaróvár and Feketeerdő (F), 2001-2003)




1. **Free pollination = no caging**
2. **Caging for the first half of the flowering period**
3. **Caging for the second half of the flowering period**
4. **Caging for the two-thirds of the flowering period**

 **There is a statistically significant difference compared to the mean values of 1.**
**(significant at P=5% level)**

*Figure 19* Number of viable seeds per fruit of apple cultivars as affected by the duration of insect pollination in the afternoon
(Mosonmagyaróvár and Feketeerdő (F), 2001-2003)




1. **Free pollination = no caging**
2. **Caging for the first half of the flowering period**
3. **Caging for the second half of the flowering period**
4. **Caging for the two-thirds of the flowering period**

 **There is a statistically significant difference compared to the mean values of 1. (significant at P=5% level)**