

# The temperament traits and the hygienic behavior of honey bee (*Apis mellifera carnica* Poll.) from Serbia

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**Abstract:** The aim of this study was to determine the temperament and the hygienic behavior of the six honey bee (*Apis mellifera carnica* Poll.) regional populations from Serbia. Three of these populations were derived from lowland area (Rasina valley, Morava valley, Banat), and three were derived from mountain area (Sjenica (Pester), Timok, Kopaonik). The observed traits were swarming impulse, aggressiveness, honey robbing and the number of cleaned cells in the 24 and 48 hours after damaging (pin-killed method). All of the studied traits were observed during the period of two generations. Each generation was tested for both years. In the first generation (mother colonies) the swarming impulse was more pronounced in the colonies of valley ecotypes. The colonies from Pešter region were very aggressive during the inspection (rated 2.5), and it was hard to handle the colonies of this ecotype. The honey robbing was especially pronounced in the colonies of Morava and Timok region. The highest number of cleaned damaged cells was determined in the colonies of Timok region in the both inspections. The differences in the aggressiveness and the number of cleaned cells after the 48 hours between populations were statistically very significant. In the second generation, in the offspring, all of the traits were somewhat highly rated. The aggressiveness in the colonies from Pešter region was rated 3.17, and is still restrictive factor for colony handling. In total, the colonies of this generation were susceptible to honey robbing (average grade was 3.61). The differences between rates of honey robbing, and the differences between the numbers of the cleaned cells after 48 hours were statistically very significant. The differences between rates of swarming impulse were statistically significant.

**Keywords:** honey bee, swarming impulse, aggressiveness, honey robbing, hygienic behavior

## INTRODUCTION

Domestic Carniolan honey bee (*Apis mellifera carnica* Poll.) populates the area of Balkan Peninsula, Panonic Basin and the south-east side of the Alps. This honey bee race is, recently, in great expansion in Europe and worldwide (Ruttner, 1992). Until now, the existence is confirmed of large number of Carniolan honey bee regional populations (Ruttner, 1988). Jovan Živanović, forefather of Serbian beekeeping, has, in the end of the 19<sup>th</sup> century, determined several differences among the honey bees from different regions of Serbia. The first trials in the differentiation of local population were done by Grozdanić in the twenties of the last century. The existence of regional populations of Carniolan honey bee in Croatia and Slovenia is described by many authors.

Domestic Carniolan honey bee has mild temperament. It is adaptable on the various climatic conditions, which makes it one of the most bred honey bee races in

the world. It winters well in the conditions of continental climate and show fast spring development.

Adam Kerle has, after studies that lasted several years, described this honey bee race as mild, resistant to diseases, with long lifespan and strong wings. Due to the increased swarming impulse, the most likely due to fast spring development, it is stated that this race is not fertile enough (Ruttner, 1992).

Poklukar (1999) states that the ability of Carniolan honey bee to winter in the weaker colonies and to have very fast spring development is the reaction to the climate conditions of the regions from which this honey bee has originated (subalpic climate of the north-west regions of Slovenia). This trait can, also, be the cause of the increased swarming impulse. The queen bees of this race stop to lay eggs in the winter when temperature drops, and starts, again, very quickly in the spring when the daytime is longer, although the temperatures are still relatively low.

This is very calm honey bee race, which is easy to handle even without special protection. These honey bees have highly developed sense of orientation. They are known to have extremely strong instinct for food gathering. This can, sometimes, be a problem due to possibility of honey robbing occurring in the autumn. The hygienic behaviour of honey bee is the natural mechanism of defence against foulbrood (Rothenbuhler, 1964) and other brood diseases (Spivak and Gilliam 1998a, 1998b).

The aim of this study was to determine the behaviour of the honey bees during the handling them. The honey bees were of different regions of Serbia. Two generations of regional populations were studied.

## **MATERIAL AND METHODS**

In this experiment were included the honey bee colonies sampled from six different localities in Serbia. Three of these localities are with higher elevation (Kopaonik, Pešter and Tupižnica near Knjaževac), and other three are of lowland type (Banat, Rasina river valley and Morava river valley). After the thorough study, three colonies were selected from each locality. These colonies were subjected to the further testing.

The colonies of Rasina region were obtained from the apiary of the Institute for forage crops, which was comprised of over 70 colonies. Samples of colonies of Sjenica region (Pešter) were obtained from Kladnica village (elevation 700m) from beekeeper Ismet Šabanović (over 200 colonies). Morava region is represented with the colonies sampled from the apiary of Milija Racković (80 colonies) from Cvetka village near Kraljevo in Morava river valley. The colonies sampled from the apiary of PIK Bečej (1000 colonies) were the example of colonies of Timok region. The colonies of Knjaževac originate from the apiary of Vukašin Milisavljević (200 colonies) from Zubetinac below Tupižnica on the location Gladna Reka (elevation 620m). The colonies of Kopaonik region were sampled in Borjanci village (elevation 800m) from beekeeper Novaković Dojčin (over 30 colonies).

The honey bee temperament was assessed in the spring and autumn inspections, in the same time as the other production traits, as was given in the Rule book of honey bee selection (Official handbook RS, 1996).

The aggressiveness was assessed by the honey bee behaviour during the inspections, did they sting and for how long they remained on the removed frame. This trait was graded 4 to 1 in each inspection (4 – very calm, 3 – calm, 2 – irritated (they move from brood combs to honey combs), 1 – aggravated (they leave the

combs)). For further selection, only queen bees marked with 4 are considered (Mladenović, 2006).

The swarming impulse was, also, graded with marks from 4 to 1, (4 – very good, 3 – good, 2 – bad, 1 – unacceptable). The swarming impulse was assessed depending on the appearance of the urge for natural swarming. For selection, only the queen bees marked with 4 are used.

The appearance of honey robbing is, as with previous traits, graded with marks from 4 to 1.

The hygienic behaviour was determined using the “pin killed” method. Using the medical needles, 100 brood cells were damaged. After 24 and 48 hours the open and cleaned cells were counted.

## **RESULTS AND DISCUSSION**

### **Aggressiveness of the honey bees**

The colonies from different regions of Serbia have had high variability (table 1 and table 2). The colonies of Rasina region are assessed as satisfying for this trait in the first and the second generation. The colonies of Pešter region were very aggressive in the first year. This made them hard to handle during the inspections, thus deeming them unsatisfying. The aggressiveness of the colonies of this region is somewhat decreased in the offspring generation, but was still a problem for handling. The colonies of Morava region had the similar behaviour as the colonies of Pešter region. The colonies of Banat region had different behaviour in the first two years. The handling them was somewhat hard due to their aggressiveness, but not as the colonies of Pešter region.

The offspring which was tested in the third year had shown decreased aggressiveness. The fifth group (Timok region) was the mildest (marked 4 – very calm bees), so these colonies were very easy to handle. The honey bees stayed on the removed combs. This type of behaviour has proven to be a problem due to susceptibility of such colonies to honey robbing. Namely, these colonies had been attacked by bees from other colonies, and had problems in the colony defence. The behaviour of the colonies from Kopaonik region was not satisfying in the first two years. The offspring of this group have shown much satisfying behaviour. By variance analysis it has been determined that the colonies from Sjenica are statistically different than the colonies from other regions.

The offspring of the selected queen bees, tested in the third and the fourth year, have shown very interesting effect. Namely, the honey bees from the Pešter region were milder than in the previous years, while the aggressiveness of the bees from the Rasina region was increased. Other groups have had satisfying temperament. Such behaviour in the third and fourth year can be explained by the fact that all queen bees mated freely in the uncontrolled conditions with various drones, so there was the mixing of genetic material between groups which could result in the decreased aggressiveness of the offspring.

Table 1. The marks of the I generation colonies for swarming impulse, aggressiveness, honey robbing and hygienic behaviour (2002-2003)

Trait	Temperament						Hygienic behaviour	
	aggressiveness		swarming impulse		Honey robbing		24 <sup>00</sup>	48 <sup>00</sup>
Region	2002	2003	2002	2003	2002	2003	2003	
Rasina	4,0	3,7	4,0	3,3	4,0	3,7	59,3	82,7 <sup>a</sup>
Sjenica	2,7	2,3	4,0	3,7	4,0	3,7	43,7	64,7 <sup>b</sup>
Morava	3,7	3,7	4,0	3,0	3,0	3,3	46,3	64,3 <sup>b</sup>
Banat	3,7	3,7	4,0	3,0	4,0	3,0	57,3	81,7 <sup>a</sup>
Timok	4,0	4,0	4,0	4,0	3,3	3,0	61,0	85,7 <sup>a</sup>
Kopaonik	3,3	3,3	4,0	3,7	4,0	3,3	50,7	71,7 <sup>ab</sup>
Average	3,50		3,72		3,53		52,22	74,80
F – test	6,27 <sup>**</sup>		0,48 <sup>ns</sup>		2,06 <sup>ns</sup>		1,04 <sup>ns</sup>	11,99 <sup>**</sup>
Lsd 0,05	0,58		0,61		0,61		18,75	8,05
Lsd 0,01	0,78		0,82		0,83		26,29	11,29

Such marks can, also, be the result of the uniform apitechnic measures, good nectar flora and good weather conditions. However, it is hard to determine how or if the aggressiveness will decrease in only one generation. In the second, same as in the first generation was determined that the colonies from Sjenica region were statistically significantly different than the other colonies.

Speaking of the defensive behaviour, or the attacking and stinging behaviour, Hunt et al. (1997; 1998) have determined that the intensity of this trait is determined genetically. Many authors give advantage to calm bees than productive bees in the selection (K r i v c o v , 2003). As with all other traits, there is the variability, not only within the specie *Apis mellifera* L., but , also, within every race of honey bees, and even within each colony.

Table 2. The marks of the II generation colonies for swarming impulse, aggressiveness, honey robbing and hygienic behaviour (2004-2005)

Trait	Temperament						Hygienic behaviour	
	aggressiveness		swarming impulse		Honey robbing		24 <sup>00</sup>	48 <sup>00</sup>
Region	2004	2005	2004	2005	2004	2005	2004	
Rasina	3,7	4,0	4,0	4,0	3,7	3,7	63,3	84,3 <sup>a</sup>
Sjenica	3,0	3,3	4,0	4,0	3,3	3,7	56,3	73,3 <sup>ab</sup>
Morava	4,0	4,0	3,7	3,7	4,0	4,0	54,3	70,7 <sup>b</sup>
Banat	4,0	4,0	3,3	3,7	3,7	3,7	52,7	76,7 <sup>ab</sup>
Timok	4,0	4,0	4,0	4,0	3,7	3,3	62,3	85,3 <sup>a</sup>
Kopaonik	4,0	4,0	4,0	4,0	3,3	3,3	49,3	71,3 <sup>b</sup>
Average	3,83		3,86		3,61		56,37	76,93
F – test	12,00 <sup>**</sup>		3,12 <sup>*</sup>		1,33 <sup>ns</sup>		1,03 <sup>ns</sup>	2,45 <sup>*</sup>
Lsd 0,05	0,28		0,36		0,57		16,70	12,72
Lsd 0,01	0,37		0,49		0,77		23,42	17,84

Many authors describe this race as very calm, very easy to handle (Kulinčević, 1966; Kulinčević et al., 1997; Ruttner, 1988; Bilaš and Krivcov, 1991; Mladenović and Mirjanić, 2003 and others). The behaviour of the second group is characteristic for this regional population that has been

described by the number of authors who have studied the honey bees of Sjenica-Pester region (Vlatković, 1957; Stanimirović et al., 1999d, 2000; Ćirković, 2002)

### **Swarming impulse**

The swarming impulse in the first year was not a problem, so entire group received the maximal mark (table 1.). Since the moment the colonies were introduced into the experiment, neither colony entered the swarming impulse. In the second year, however, swarming impulse was problem for the most of colonies in the experiment. The period after the fruit blossoming and during the blossoming of false acacia was characterised by heavy rainfalls and warm temperatures. The number of young honey bees was increased, and due to inability to fulfil their tasks, (because of weather conditions at the time), the swarming impulse occurred.

Only in the colonies from the Timok region the swarming impulse was not a problem. The colonies from the Banat and Morava region were deemed the worst for this trait. Besides the weather conditions which could induce the swarming impulse, it did not occur in some colonies. This was the case in all three colonies from the Timok region. However, for all colonies, except for the one colony of the Rasina region, the absence of swarming impulse can be contributed to their low condition. In the colony from the Rasina region this could not be the reason, due to its high strength in the spring, so this behaviour can be contributed to genetics.

In the third year (offspring) the swarming impulse occurred in the colonies from Morava and Banat region (Table2.). The colonies from other regions have had no problems with this trait, and so were marked with maximum marks (4.0 – very good bees). In the fourth year, the colonies have had nearly identical marks as in the previous year, so the total mark for this year was quite high (3.9). In this year, queen cells were present in the one colony from Morava region and in the one colony from the Banat region.

The swarming impulse is, also, increased due to human interference, due to earlier practice in the selection to favour the bees which had the increased swarming impulse, for the easier reproduction of the bees (Krivcov, 2003). By the invention of the hive with mobile frames and the methods for the artificial swarming, the natural swarming impulse became unwanted trait and I considered making great economic damage to the modern beekeeping.

Билаш и Кривцов (1991) claim that the occurrence of the swarming impulse in the Carniolan honey bee depends on the availability of the supporting nectar plants in the spring. The Carniolan honey bee has high tendency to the swarming, but this trait can be manipulated through selection. Swarming impulse is lower in the Carniolan honey bees from Serbia, than in those of Slovenia (Kulinčević et al. 1997). Mladenović and Mirjanić (2003) claim that the Carniolan honey bee does not overly tend to the requeen while the swarming impulse can be significantly decreased using the corresponding selection program.

### **Honey robbing**

In the first year there was clear difference between studied populations for this trait. The best mark for this trait was given to the colonies from the Rasina, Pešter, Banat and Kopaonik regions (table1.). The colonies from the Morava region attacked other colonies very often, while the colonies from Timok region were usually under

the attack and had the problem to defend the colony. In the second year, honey robbing was common especially at the end of the summer and during the autumn.

The colonies of all groups were involved in the honey robbing, whether they attacked or were under attack. Especially the colonies from Pešter and Banat region were active in the attacking other colonies. One colony from Timok region is lost to honey robbing.

In the third year, colonies shown satisfying results, except the colonies from Kopaonik, which were often under attack (table 2). In this year, the colonies from Morava region received maximum marks. In the fourth year, the colonies from Morava region received maximum grades, while in all other groups were colonies graded with lower marks. The problem was in the colonies from Kopaonik region due to inability of those colonies to effectively defend the nest.

Билаш и Кривцов (1991) claim that the Carniolan honey bee has the lower instinct for honey robbing than Italian honey bee race, but higher than Caucasian race. The Carniolan honey bee colonies are capable to defend the nest from foreign honey bees. Kulinčević et al. (1997) claim that the honey bees of this race have very good instinct for orientation and the honey robbing instinct is not too present.

Somewhat different is stated by Poklukar (1999), who claims that this race have very strong instinct for food gathering, which can be a problem in some cases. Especially, this trait can have negative effect in the autumn, when it can lead to the honey robbing between colonies if the beekeeper is not careful when feeding the colonies.

### **Hygienic behaviour**

In the second year, besides the all other traits studied in the last year, the hygienic behaviour was also studied. In the first trial, the colonies have cleaned 52.22% of cells (table 1). There were great variations between different regional populations. The most cells were cleaned by the colonies from Timok region, and the least by the colonies from Pešter region. In the second trial, 74.80% of cells were cleaned. Again, the most cells were cleaned by colonies from Timok region (85.7%), while the least were cleaned by colonies from Morava region (64.7%).

Using the variance analysis, it is determined that the origin have significant influence on this trait in both the first and the second period. Colonies from Kopaonik were not significantly different, while other regional populations were classed in the two separate groups significantly different from each other. In the first group were colonies from Rasina, Banat and Timok regions, and in the second were the colonies from Pešter and Morava region.

In the third year (offspring), in the first study term, average 56.37% of cells were cleaned, which makes 4.15% more than in the last year. Best in this period were the colonies from Rasina and Timok region. The colonies from Banat and Kopaonik region have cleaned fewer cells than last year.

This honey bee trait is very variable, and due to that there are different results of different authors. In the studies of Mladenović et al. (2005), it is stated that the honey bees from West Serbian regions cleaned 41.46-47.37% of damaged brood cells in 24 hours. In 48 hours after the damaging, they cleaned 57.15-65.75% of cells. Pejović (2001) determined in his studies that the colonies from Rudnik have more expressed hygienic behaviour (93.6%) than the colonies from Pešter (91.50%) and from Mačva (91.45%).

## CONCLUSION

Based on the results of the study of temperament and hygienic behaviour of colonies from six Serbian regions next can be concluded:

- Occurrence of the swarming impulse was more influenced by environment than origin of honey bees. Weather conditions in spring period, before and during the false acacia blossoming, had great influence on the occurrence of swarming impulse.

- During the four-year study, the aggressiveness of the honey bees varied. The colonies that were very aggressive, in the beginning, became calm, while certain calm colonies became more aggressive.

- The honey robbing depended mostly on the strength and aggressiveness of the colonies, meaning that the weaker and calmer colonies had problems to defend the nest.

- The origin of the honey bees had the significant influence on the hygienic behaviour of colonies. The colonies of Timok region had the best results for this trait in the entire study.

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