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## VARVOACIDAL EFFECT OF FLUMVAR AT CONTROL OF HONEY BEE VARROASIS

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#### ABSTRACT

**Aim:** Preliminary evaluation of flumvar efficacy in control of varroasis in some Tehran province's apiaries, Varroasis is the most important parasitic disease of honey bees that in addition of direct damages due to perforation of honey body can provide transmission the kinds of viruses and bacterias. However, during recent decade were carried out the efforts in order to biological control of varroasis but the main solution against varroa is chemical control by pesticides yet.

**Methods:** In this study 30 hives in 2 experimental group's induding negative control, and flumvar were surveyed. After determination of primary in festation in experimental group with flumvar was applicated according to theirs manafactere subsequently, 40 days after treatment the varroal in festation were determined again.

**Results:** flumvar was decreased varial infestation from 12 to 2 and 11 to 1% respectively. The efficacy of flumvar was 90.26% respectively. No side effects were seen after flumval application. According to the more than 90 percent of flumvar efficacy the variacidal effects of this pesticide is suitable.

Key words: flumvar, varroasis, efficacy, apiary, Tehran.

## **INTRODUCTION**

The life of a bee is one of the wonders of life and one of the secrets of creation. Throughout its short life, this creature is engaged in the activity, production and protection of the hive, ie its habitat. Man uses all bee products in some way and tries to force him to produce more with higher quality. This creature is important due to the production of honey, wax, pollen, propolis, venom, royal jelly and hives and pollinators.

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In the last century, beekeepers in the world have been as extensive as other agricultural activities and include hundreds of thousands of colonies with new technologies. In the villages of some countries, people still work side by side with beekeeping to earn more money in addition to their main agricultural work. The main income of beekeeping in developing countries is the sale of honey and wax, and some beekeepers also raise queens and produce envelopes. Therefore, this small insect is the source of services and positive effects for humans, agricultural products, the environment, forests and pastures.

The economic benefits of bees determine the importance of fighting the disease, and recognizing the diseases and pests

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that threaten the insect is very important (3).

The most important of these are the Acarapis and Vedi tropilaeipa Clara mats. Varroa disease is one of the most dangerous diseases of bees, which is caused by Jacobsoni mite varroa, and this parasitic disease can destroy bees in a short time and cause a lot of damage. Due to the transfer of bees from one region to another, this disease has been widely spread and has been mainly due to the purchase of the queen and the transfer of mites.

Varroa Jacobsoni is probably the largest Asian mite, and in Iran there have been reports from various parts of East and West Azarbaijan provinces that the mite has been planted by the Ministry of Agriculture. Studies have shown that bees in most parts of the country have been infected with this mite and have damage. Currently, caused in our country, various therapeutic methods are used to combat this disease and prevent its spread, the most important of which is the use of chemical compounds produced by various and reputable international companies, and some of these chemical compounds have had significant positive therapeutic effects and most beekeepers are satisfied with their effects.

**Varroasis, dangerous bee disease** Varroasis disease is caused by a mite named Varroa Jacobson. Mite Varroa is subordinate (Acarina), a parasitic order below Gamasida (Mesostigmata), and the Varraoidae family. This parasite is a parasite of larvae, pupae and adult bees and is therefore considered a great danger to the beekeeping industry (8).

**History of Varroasis in the World** Mite Varroa was first reported under this name in 1904 by Oudmans on an Indian bee in Java. In the same year, it was described and classified by Edmonds. In 1909, it was

reported under another name by Suzuki in Japan. In 1951, on the islands of Singapore, a collection of Indian bees was collected and named after Mir Mozur Kon Ridi Ganter was named. In the first fifty years, from 1904, when it was recognized, until 1953, there were limited reports on it, until it was first reported in 1953 in the eastern part of Russia, followed by Japan and China. In 1975, Obama first photographed the mite, which had been collected from an Indian bee. That same year, Kishida saw Mite Varroa on a European bee in Japan. In 1962, Dolphinado observed and collected mite varroa in Hong Kong on European bee infants (10). In 1971, colonies of European bees that had been damaged by varroa outside Asia were identified (7). In 1975, Akratanakul and Burget warned in articles that Mite Varroa might one day become the most important bee parasite in the world

(6). In 1983, the life cycle of Mite Varroa was described by Ifantidis. A study in Brazil shows how mites spread from beginning to end. In this study, 520 colonies from 26 beekeepers were selected. All of these colonies were infected. However, a year before the survey, Mite Varroa had not yet been reported from the country (7). At present, this mite exists on all continents of the world except Australia and is expanding and settling further (6).

History of Varroa disease in Iran The report of Mite Varroa's arrival in Iran is almost contradictory. According to Ms. Eva-Kerin, in 1978, a number of beehives in Iran were destroyed by contamination with Mite Varroa on the shores of the Caspian Sea. Tirgari contacted the author of the above article and declare that the report on the entry of Mite Varroa into Iran in 1978 is invalid. Professor Ratner says that in 1981, he observed a sample of Varroa mite from the Azerbaijan region, and this is the only accurate and reliable news that this bee is infected with this mite in Iran.

In the fall of 1985, the beekeeping of Iran, which is caused by Maita Varroa, was severely damaged. According to Ratner's remarks, the distribution of mites and their damage to bee hives in Iran, the date of infection of bee hives in Iran can probably be considered as 1979-1980. Because if Mite Varroa arrived in Iran earlier than this, considering the migration of bees and the non-use of any Mite drug to reduce this pest, in some regions, especially East and West Azerbaijan, Mite harmed the hives earlier than 1995-1994 in bees. In any case, in August 1984, the National Livestock Organization officially announced the existence of mites in Iran (10).

Rahbari *et al.* (2016) studied the distribution of mite varroa in Iran and reported the percentage of contamination in different regions. They noted that the route was from Turkey and Russia (7).

The statistics showed that the primary source of contamination was the provinces of East and West Azerbaijan, and then, due to migration, buying and selling of hives, this mite quickly spread other parts of the country. to Observations in 1985 indicate heavy casualties in Khorramabad, Lorestan. During a visit to 25 beehives with about 3,200 hives, 2,000 hives were lost and the remaining hives were so weak that they were unlikely to survive the winter. Meanwhile, the percentage of pollution of Lorestan hives to the provinces of Azerbaijan and Hamedan has been lower. Statistics show that the average damage to the hives of beekeepers which migrated to Khuzestan was 40%.

Considering the total number of hives in Iran in that year, which was one million colonies, it was estimated that about 41.3056 colonies of honeycombs were destroyed by the Mite Varroa attack in the fall of 1985. Today, colonies infected with mite varroa are found in all parts of the country (9).

Due to the history of bee colonies in the country, due to the unfamiliarity of beekeepers with prevention and control methods, a large number of hives are destroyed every year throughout the country due to severe pollution and cause great damage to beekeepers and the beekeeping industry. In genkeral, studies have shown that if the mite population in the hive is higher than 20%, the disease and its damage to bees will be evident, but if it is less than 20%, it is tolerable for the hive (12).

## Mite Varroa morphology

This mite has a three-part body, the first part is called gnathosoma and the second and third parts are called joint and idusoma. The nature of the mature egg is flat, 1.1-1.2 mm long, 5.1-6.1 mm wide and reddish brown in color, and can be seen with the naked eye. (5).

The male mite is triangular in shape and has a sharp body on the front and is smaller than the female. It is 76.0 mm long and 71.1 mm wide. The mite is light yellow to light gray. The size of the legs in males is longer than in females. The posterior plate of the body of the mitochondria covers almost the entire body surface and the oral parts of the gonatosa are hidden beneath it. Gonatozoa has a pair of chelicera and a pair of pulp pads. In males, the Schleser is modified to transport sperm and is not fed. Therefore, the male cannot feed on adult bees and will soon die after mating. The body of an adult bee is mating only with female mites (9).

In the middle part of the mouth there is a structure called the hypostome. The suction operation is performed by the pharynx. The toes of the mites' legs are wind-blown, so they can easily attach to the bee's body. In addition, the hair on the surface of the abdomen is hardened and able to adhere to the hair of the bee's body, so that the bee is unable to separate it from its body (6).

In general, the size of the Varroa mite varies, and this difference in size can be seen even in samples collected from a hive. The size and shape of the mites on the body of the European bee and the Indian bee also vary. Racial and environmental factors are likely to be involved (10).

In general, the size of the Varroa mite varies, and this difference in size can be seen even in samples collected from a hive.



Picture 1-2 Mite Varroa Jacobsoni

#### How to publish Mite Varroa

The population of Mite Varroa in a hive depends on the growth and developmental period of the bees in that hive. Varroa mites spend most of the summer on larvae and bees. (6). If the population of mites in the hive is higher than 20%, the disease and its damage to beekeepers are evident. But if it is less than 20%, it is tolerable for hives. (7)

The publication of this site is as follows:

- Transporting hives from one point to another
- Transportation of hives in migratory beekeeping
- Flying worker and male bees in beekeeping areas
- Baby hives that run away from hives.
- Buying and selling bee colonies and transferring them among beekeepers
- Buy and sell bee queens

Ranter and Ritter reported in 1980 that mite varroa is naturally released about 2 to 5 kilometers into Germany each year.

Otis *et al.* (1981) reported that the rate of spread of mite varroa in African bees in South America was higher because these

bees and their hives could fly up to 131 km per year (6).

Damage caused by Varroa to bees Today, Mite Varroa is considered as a very important bee parasite in the global beekeeping industry. This mite was originally a parasite of the Indian bee and, while in contact with the European bee in Japan, the Far East and the former Soviet Union, it has been able to live well on it. Mite Varroa has been able to come to terms with his main host, the Indian bee, over the years, and has not done much damage. However, for European bees, this mite is important and can cause irreparable damage because, unlike Indian bees, European bees do not effectively defend themselves against mites.

European bees do not separate Varroa mites from each other's bodies, while Indian worker bees do. Temperatures in European beehives are closer than the temperature in Indian beehives to the desired Varroa temperature. European bee hemolymph has a high level of young hormone, which is good for growing mites. The number of male bees in the European breed is higher than that of the Indian bee, and the female varroa lays her eggs in the cells of the bee baby, but prefers male infants to the female. For this reason, it suffers much more from the damage inflicted on their colonies by Varroa (5).

Throughout his life, Mite Varroa feeds on the hemolymph of newborns, pupae, or adult bees (workers, males, rarely queens). Mite Varroa chooses soft areas of the body to feed on bee hemolymph. For this purpose, it is often placed between the loops of the body, especially the first loop, between the head and the chest, and between the abdomen and the chest, and feeds the bee's hemolymph by entering the chelicera (6).

Varroa also injects a substance into the host that can cause anesthesia and is anticoagulant and digestive, which endangers the life of the bee. As a result of this feeding, bees may not be much harmed, but the presence of wounds in different parts of the body can cause pathogens such as chronic leukopenia, neonatal waxworm and fungal diseases that complicate the situation and make it difficult to control the parasite (3).

Mite prefers male cells to worker cells, which prevents the male bee from mating, so when the baby is born or the colony changes its queen, the queen does not mate and the colony is doomed. Or the fertility of its queen is greatly diminished (7).

In the first year of infection, a colony is often between 1 and 10 mite in a hive, and in the second year it reaches 100 and in the third year it reaches more than 1,000. In this case, there is no deficiency in honey production and no clinical symptoms. In the fourth year of infection, due to the increase in the population of mites in the hive and severe feeding, incomplete bees are observed in the hive. These bees have deformed legs and short bellies and wings. (6).

The characteristic of varroasis is that it is spread by adult female mites, and male and infant mites and parasitic nevi are inside the closed cells of bees and feed on hemolymph larvae and bee pupae. The amount of hemolymph that this mite receives varies from season to season, but the maximum is 41% of the body weight of the mite and 120% of the weight of the bee's body. As a result, bees that lose their hemolymph are unable to survive, and their lifespan is shortened.

The normal lifespan of a worker bee is 180 days in winter and 40 days in summer. The lifespan of a worker bee is reduced to 18 days if there is a mite on it, to 18 days if there are 2 mites on it, and to 8 days if there are 2 mites on it, and to more than 2 mites if it exists. On the bee's body, it lasts for several days.

In general, in colonic contamination by Mite Varroa, we will have colonic weakness and subsequent colonic destruction. When colony infestation reaches 30 to 40 percent, bees leave the hive and die outside the hive, causing great economic loss. Also, following the decrease of bees in the hive, infant care is not good and after a while, the colony is destroyed (7).



Figure 1: Mite Varroa on the pupa

**Symptoms of Varroasis veins** Usually, clinical symptoms are not seen because the level of infection is low in the early stages of infection. However, in the third to fourth years, the damage is severe. These symptoms include:

Behavior change the bees on the bee's body sometimes change their location on the bee's body to suck more hemolymph. As a result of this movement, bees behave abnormally by walking fast or fluttering their wings.

If the mites are placed in close proximity to the bee's wings, the bee loses its ability to fly. Mite Varroa uses the blood or hemolymph of bees and will cause defects in the bees, so that the bees born and infected with mites are smaller in size and these bees have deformed legs and short bellies and wings. Such bees are unable to perform their duties and rarely survive. Due to feeding on bee blood, we have colonic weakness and subsequent death of bees.

Sometimes infected larvae may have abnormal movements and even leave the cell and fall to the hive floor. If there is a lot of mite contamination in a room, it will cause the death of larvae and pupae. If the number of dead larvae and pupae is small, the worker bees pull them out and clean the cells, but if there is a high mortality rate, the bees are neither able nor willing to clean the cells. In this case, the dead larvae and pupae in the cell will look like American locusts, and the walls of the cells will turn white from above.

If there is more than one mite in each chamber of the pupa, severe damage will be done to the pupae, and if more than 6 mites of the blood of one pupa feed inside the cell, the pupa will lose a lot of weight and this pupa will enter the cell or after leaving. The cell will die. If the infected pupae are used by 2 mites, their lifespan will be short and they will live only 9 days after birth. These bees are unable to hibernate.

Male bees are weakened due to mites and lose their ability to fly and mate, and the queen of infected hives often does not mate or their mating is incomplete, so in addition to being parasitic, the hives produce incomplete queens or pairs. They have not been caught and in this way they are causing damage. Infection occurs as a result of bite bites, infections, and secondary diseases, and the physical strength of the bee is reduced by a decrease in blood volume and protein. The severity of some diseases depends on the cause of the infection, such as European leukemia and cases of neonatal disease.

In severe mite infestation, the infestation is irregular and the spawning state is similar to that of the old queen in the hive. Adult bees infected with a mite will reduce their lifespan by 150% (9).

#### Ways to diagnose pollution

Contamination should be diagnosed in the early stages because if several years have passed since the infection (3-4 years), although varroa mite can be distinguished from its appearance, it will be difficult to treat mite due to colon weakness and severity of infection. There are several ways to diagnose an infection, each of which is effective at a particular time, and using them at a particular stage of the infection will have successful results among these methods.

## Non-chemical methods Examination of hive floor particles

Many mites will die after leaving the cells and fall to the hive floor. This method can be used for hives that have no history of infection but have suspicious mites. In the fall, when the bees gradually stop flying out of the hive, they cover the bottom of the hive with white paper and place a net frame on the paper plate. The diameter of the mesh holes in the base frame should be 3 so that the bees cannot come in contact with the paper plate and therefore the possible mites. In the spring, after removing the larvae from their cells, the paper is taken out and searched for mites in the rubbish dump. The mites can be separated from other debris by floating. For this purpose, all debris is first poured into 95% alcohol and then into 50% alcohol. The mites float on the surface of the solution and can be collected (6).

#### **Examination of larvae and pupae**

In this method, we open the head of the closed cells of the larvae of the last age or pupae and carefully examine the larvae or pupae or the empty cell. Mites can be detected on a 13-day-old worker pupa and an 18-day-old male pupa. The presence of white spots on the walls of empty cells is the reason for the contamination of the hive with mites. At least 100 cells containing pupae or larvae should be examined for bee and mite infestation. Because varroa mites prefer male larvae, especially on the sides of the frames, it is effective to visit and examine these cells that have male larvae (10).

#### Visiting and testing adult bees

This method is used when colonic birth is stopped or minimized. In this method, 500-1000 bees, preferably young bees that have just come out of the cell, are immersed in boiling water or water, which is some detergent, usually soap scum or dishwashing liquid. We are pouring. After shaking several times, if there is a mite, it can be separated from the bee and can be seen at the bottom of the container. Using 70% ethyl alcohol or gasoline instead of water increases the reliability of the test.

Adult bees can also be placed in a widemouthed glass and a small amount of ether added to it. In this method, adult bees are not killed and if there is mite, it is attached to the glass wall and is completely clear. In this method, bees can be returned to the hive after examination (6). Sugar Powder Method: Sugar Powder Method is a traditional method. In this method, we spray powdered sugar on the bottom of the colony and then remove the mites that have fallen off the tray or something like the bottom of the hive.

## Observing the side effects of clinical symptoms

In this method, the damage caused to pupae and adult bees is investigated and the infection is diagnosed by evaluating them. The presence of larvae and pupae and incomplete bees, adult bees with short and twisted wings is a sign of the presence of mites in the hive and contamination. However, this method is not suitable for diagnosing high pollution because at this stage the infection is severe and the colonies are weakened and there is no economic treatment.

In this method, we put a number of adult bees (about 100) in a net cage and keep them in an oven at 46-47 degrees Celsius and a relative humidity of 20-30% for 10-15 minutes. We put white paper under the cage and so we can count the spilled mites (7).

### **Chemical method**

This method uses a mite killer to detect contamination. This method can be used in field and laboratory.

#### In Field

In this method, by spreading paper on the bottom of the hive and using mite drugs to reduce the specificity of combating this mite, it is possible to study and collect mites. When using mitochondrial drugs, if there is honey in the hive, it should be removed from the hive beforehand to prevent contamination.

#### In the laboratory

About 100 live bees are placed in a net cage and the white cardboard pieces are placed in the cage. Any available chemicals can be used to remove mites from the bee's body. Papers collected on paper can be counted (10).

### Prevention

To do this, the distance between the apiaries must be increased so that the distance between them is 10 km or more so that the bees of the adjacent apiaries cannot come in contact with each other and the varroa mite is transferred from one apiary to another. Of course, the result cannot be 100%, because male bees sometimes travel longer distances and can therefore penetrate and infect the hive, which is 10 km or more away, but this has been done so far and the bee has caused a lot of pollution. It is easily transmitted, so the speed of propagation is much slower, and in many cases it is not even propagated at all. Care should be taken when buying bees and avoid buying infected populations (13).

#### Varroasis disease treatment methods

Various methods have been used to combat Mite Varroa, which can be summarized into two categories: chemical and non-chemical:

#### **Chemical methods**

One of the most effective methods is controlling of Varroa. Before using any chemicals, the colonies should be carefully examined, the hive floor should be cleaned, and all the holes and crevices in the hive should be closed. If the hive door does not close completely, place a plastic sheet on the inside door of the hive and then place the outer door on it. Sometimes it is necessary to remove one or two frames from an infected colony to make room for chemicals.

The best way is to fight with chemical poisoning. The best time to fight chemical pesticides is in the early evening or early morning, when all the bees are inside the hive. The fight against varroa mites is carried out in autumn or early spring because at this time the number of bee infants is minimized and the full mites are removed from the bee cells and are on the body of the full bee and can be killed with mite-killing chemicals. In some cases, it is necessary to remove the closed larvae, especially the new larvae, before the fight. Drugs should not be used at all in the colonies of bees during the flow of natural nectar and honey storage (6).

#### **Disinfectants (detergenty)**

Use of smoke from burning plant parts Such as: mint, eucalyptus leaves, wild thyme, thyme (12).

### Use of smoke fakes:

These substances are usually associated with potassium nitrate. The following groups can be mentioned:

Danny Krupper: This combination is the same as the Tetraditone machine. A piece of 80 g cardboard cardboard, which is enough to treat 50 hives, in a solution containing 0.75 g4-5 and 4 and 2 - tetrachlorodinthoate sulphone and 0.25 g-0.0 (ethoxy1-carboxy1) dimethyls 1,2-bis phosphoralithoate, We put. Then we put the cardboard in a smoke that contains a few bright and red coals to catch fire. When the smoke comes out, we blow it out three times with smoke. This operation should be done 2 to 3 times at a distance of 5-10 minutes. In practice, it gives the beekeeper the opportunity to smoke 50 hives at a time and return to the first hive arrives. The treatment should be repeated 2-3 times at intervals of 5 days. The bleaching property of this compound is reported to be 97%. (10)

**Varostan:** According to 6-methyle-2/3quionxaline dimetyl cyclic carbonate, it is in the form of capsules that ignite it and enter the hive through the flight hole. Due to its toxicity to infants and adult bees, it causes severe casualties.

**Phenothiazine:** One of the oldest materials used in the fight against mites. It is in the form of tablets or strips and is consumed in the amount of 1.5 grams in smoke, and all the pores, including the flight hole, are blocked for five minutes. On average, it destroys 80-70% of mites.

Frequent use of phenothiazines can cause resistance to varroa mites. In addition, in some cases, the queen is killed (10).

**Varroa:** Can be consumed in the summer in the form of smoke, but it should be consumed sparingly because it will kill bees and are harmful to humans. This drug is a carbamate (1).

**Fumilate** (**A**, **B**): In the form of smoky strips, in infant cells without cells, the closed head of the infant had 100% mite and in hives with closed cells, up to 5.93% mite (12).

**Galcron:** It is in the form of paper or cardboard. According to Smirno and Tecerno in 1976, the amount of paper tape used was one colony or one cardboard strip for eight colonies. The fight takes place three times a week (6).

Calmness: It is in the form of a flue strip that is made of potassium nitrate and amitza. It is repeated three times in the fall, 5 to 7 days apart.

**Calcutta:** Pieces of cardboard containing 0.5 g of calcaneus have been used in smoke in the laboratory against White Varroa and good results have been obtained. Due to the high durability of this toxin, there is a risk of contamination of honey and wax (9).

Varroasis veins: In the form of smoky strips of potassium nitrate with 1 to 2 ml of the active ingredient of the varroa vaccine (7).

## **Evaporative or sublimation materials**

Evaporative materials can be used effectively and without disturbing the hive and against the varroa mite. These materials are liquid or solid, and the vapor or gas emitted from them has the property of mimicking and poses less danger to humans. By placing them in the hive for two to three weeks, the drug reaches all the available mites and kills them.

**Formic acid:** In some countries, the use of formic acid (ant ant) has become common against Varroa mite. Formic acid is marketed differently, but ferric acid solutions with varying degrees of purity, including 65% and 90%, are available in most countries (Van Wayne, 2008).

Formic acid is used to transform the hive, so that it gradually cools and causes the mites to die. For an average colony, a total of about 80 to 120 milliliters of 65% formic acid is consumed. For this purpose, 20 to 30 ml of 65% formic acid is poured into a glass or plastic bottle container and a cotton ball is placed in its entrance or cleaned with a layer of cloth and placed on the bottom of the hive to be gradually cooled. This is repeated four times, four days apart (6). Another advantage of formic acid is that its vapor can penetrate into the closed cells of the nostrils and cause the death of mites.

The use of 85% formic acid in the amount of 50 ml resulted in 100% mite mortality (12).

**Chlorophenol:** It is in the form of a strip and is placed between frames 1, 2, or 9 and 10 of the hive, which are slightly open. One bar is enough for one colony. Close the flight valve for 30-60 minutes and after 1-2 days, collect and destroy the mites that have fallen on the hive floor. This combination is effective when the ambient temperature is between 15 and 18 degrees Celsius (6).

**Thymol:** This drug is in the form of crystalline powders or a solution in vegetable oil. Timol is one of the first effective drugs to control Varroasis veins, which has been used in Eastern Europe. Its effect is about 80%, which of course depends on the amount of evaporated thymol (5).

**Sulfur Combination (K2S2O5) Dithioneates:** This compound slowly releases sulfur dioxide into the hive air. It has about 80% mite effect. It also stays in the hive for 25 to 47 days (7).

**Spray materials (liquid spray) Prizine** (**Coumaphe):** This mite extract contains 2.3% of the active ingredient DD-diethylthiophosphate and 3-Choro-4methylumbelliferone and dissolves the prismine emulsion in water in a ratio of one to fifty and 50 cm3 of dilute solution. It is placed for a hive.

Amitraz: This drug contains 5.12% of the active ingredient. Amitraz is one of the formaldehyde mite killers that has been converted to the effective substance N-demethylchlordimeform after application and is effective on all stages of mite life.

The amount produced to be applied to the bee solves one milliliter of amitraz in 10 liters of water and sprays 80 to 250 (depending on the size of the colony) milliliters on the bees. It should be noted that amitraz is toxic to bee infants and therefore should be avoided by spraying frames containing infants with this compound.

This material can be prepared as cardboard strips in the dimensions of 9.5 2 2.5 cm from filter paper and soaked in a solution of potassium nitrate or sodium, and then allowed to swell, and then each strip is sprayed with 0.1 ml of amitraz solution.

**Azantol:** Its active ingredient is coumafus and in Greece it has been 81 to 100 percent effective against varroa (7).

**Apitol:** 2 (2, 4-dimethylphenyl-imino) -3methyl-4-thiazoline-hydrochloride is an effective ingredient. Dissolve 150 grams of sugar in half a liter of lukewarm water and then dissolve the contents of the package in 10 grams of apitol. From the prepared solution, 50-100 ml (depending on the size of the colony) is spread evenly between the shoots and on the beehives. It is recommended to repeat the spray one week apart (6).

**Sucrose:** The active ingredient in this medicine is octanovate stress sucrose. This medicine is a liquid diluted in water and should be sprayed between the beehive frames. This drug causes the varroa to suffocate, or the wax on the varicocele cuticle to dry out, causing the varroa to dry out.

In a study conducted by Stinglin *et al.* They reported a 3.75% reduction in mite varroa using the drug. The duration of treatment with this drug is 7 to 10 days (5).

**Spray or spray powder** Thymol: In Russia, the use of 0.25 g of thymol powder in frames has caused 73 to 98% of varroa losses. It may also reduce the reproductive power of mites in closed cells. If this substance is used

as a cooling agent, it will have a very small effect on the mites.

**Malathion:** Powder containing 0.5 to 1 percent of Malathion is sprayed 1-2 times a day in the hive for 7 days. In Greece, this toxin has been shown to be effective against varroa and has had no adverse effects on adult bees and infants. The recommended dose is 4-5 times and two days apart. It is time to consume the poison of winter and the time when a baby in the hive of existence or its amount has reached its minimum (10).

Cinnacar: It is in the form of powder and the content of sugar and various chemicals such as 1.5% (chlorine phenylate, bromopropylate, etc.) and 0.3% Tadion.

Consumption is 50-100 grams per infected colony per hive population. It is consumed 2 to 3 times in spring or autumn. The spraying interval is 10 days in autumn and 15 to 20 days in spring. In Romania, up to 90% of this drug has been effective, but its use in Germany has not had much effect on mite (9).

**Sulfur:** In Thailand, the use of sulfur in the fight against Varroa mite has reduced the number of mites in the hive. Also, the use of a 1: 1 mixture of sulfur and naphthalene has been effective in the fight (10).

Herbal powders sprayed on frames such as: Romero, Penny Royal, Garden Sage and Thyme (7)

**Oral substances or solutions** These substances are fed to the bees as a solution in water or syrup and enter the bloodstream of the bees through the gastrointestinal tract.

**Galicron:** This drug is also called chlorodimeform hydrochloride. 35 ml of aqueous solution of 0.7% of it is poured into 50 liters of water and in the fall on two occasions, it is given to bees for seven days. One of the advantages of this material is that in the cold season and without disturbing

this material, it can be done in the cold season without disturbing the bees. As soon as the mites were fed from the blood of the bees, they separated from their bodies and fell to the hive floor. It is best to cover the bottom of the hive with oil-soaked paper to trap the mites that fall to the bottom of the hive (10).

P.N: It is consumed as a syrup and its mitekilling effect lasts up to 7 days. For each hive, 0.75 to 1 ml is recommended (7).

**Herbal foods:** These include eucalyptus, fennel, origany, dates, pollen, and pollen mixed with 1: 1 syrup, such as thyme and mint (7).

## **Contact drugs**

Apistan: This compound is in the form of plastic lights and is one of the oldest artificial groups. The active ingredient of this medicine is fluvalinate. To control the Varroa mite, two Apistan strips are usually placed in a medium hive between frames 3 and 7-4 and 8. These strips remain between these frames for 6 to 8 weeks and are then removed.

The bees come into contact with these strips as they fly or move and their bodies are stained with fluvalinate, which they pass on when they come in contact with other bees.

In a study conducted by Ludosani in northern Italy in 1993, the average effect of treatment with apistani was 44.5%, with results showing the resistance of varroa to fluvalinate. However, in an experiment conducted by Cabaras and his colleagues in Italy, the effect of this drug was announced to be 99% (5).

**Gabon:** This compound is in the form of wood strips with a polymer coating and each strip contains 1.5 mg of acrine, which is the active ingredient of this drug.

How to use this combination in hives with ten beehives is to hang 2 strips symmetrically between the frames (in hives with an average population of one bar) for example between box 2 and 3 and boxes 5 and 6, then these strips. We connect to the fork.

The strips should be placed freely between the frames, so that the bees are in contact with both sides of these strips. These strips should stay in the hive for 24 to 30 days.

The bees' bodies make the most contact with the acrine, which is then transmitted to other bees by contact.

**Bayvarhol:** This substance is an effective mite killer from the group of artificial pyrethrides and is in the form of a plastic strip and its effective substance is flowmeter, which is 3.6 mg per strip (6). For each medium hive (10 population frames) four strips are recommended at a time. These strips hang between the middle frames of the hive. The strips are hung for 4 to 8 weeks and then removed. Its mitigation effect was 95%.

#### Amounts and methods of consumption

Flumevar: Flumetrin brand for fighting varroa destructor in bees. The active ingredient in Flumvar is Flumtrin. Mite Varva is a foreign parasite in the bee that affects all stages of the bee's life. By sucking the blood of adult bees and infants, this parasite reduces appetite, weakens, and reduces their lifespan.

Flutrin is one of the synthetic pyrethroids that prevents the proper transmission of nerve messages and ultimately destroys mites by disrupting and increasing sodium permeability in nerve cell membranes. The activity and movement of the bees in the hive causes their body to come into contact with the flumer tape and be impregnated with a small amount of flumine.

The movement and collision of bees with each other also causes the gradual spread of fluorine in the hive. In this way, the parasites on the body surface of other bees are also exposed to this substance and die. These strips are made to act like a sponge, and the active ingredient is gradually replaced at the point of contact with the bees, so this product is effective for several weeks. Polyethylene: It is used as a background material for plastic matrix strips in the preparation of this product.

**How much and how to use:** This product is quick and easy to use. For every 5 frames covered with bees, it is better to put a strip inside the hive.

This product should be placed in the hive for at least 45 days. It is necessary to place two strips inside the baby's room (one strip between molds 3 and 4, one strip between frames 7 and 8). Inside the small colony (Nucleus), it is necessary to place only one strip in the space between frames 2 and 3. In the case of bee envelopes, a strip of the main hole should be hung.

**Package shape and type**: This product, in the form of white plastic strips with dimensions of  $2 \times 28 \times 192$  mm, contains fluorine. Each of these 20 or 50 strips is packed in a polyethylene bag. Finally, the polyethylene bag is placed in a cardboard box. The weight of each strip is about 0.4-12.9 and each strip contains 23 mg of active ingredient.

Complete product formulation with the name of the amount and role of each of the active ingredients and additives: Performance concentration formula (%) Flumterin% 0.3 Antibacterial Polyethylene with a low density of 100% of the material of the plastic matrix of the strips

This product has no side effects and does not cause death, reproductive disorders or other disorders in bee colonies, even if prescribed for 6 consecutive times.

The interval between the last prescription of this drug and the collection of honey is at least 30 days. The effectiveness of treatment with this product is reduced in the presence of a large number of infants (more than 8 frames covered with the baby in each hive) or the excessive presence of L5 infants.

Treatment with this product should not be done more than twice a year for two consecutive years. (It is necessary to change the active ingredient in a rotating manner. When carrying this product, protective gloves should be used. Contact with this drug as well as gloves contaminated with this product should be avoided with the face, eyes and skin. Contaminated clothing is required. Wash with soap and water.

Flumvar, a plastic strip containing flumerine, is designed so that the active ingredient is released slowly and in the hive for 45 days. The most commonly used phloem in this product is about 95% effective and has high stability and biosecurity for use in beehives. This product leaves the least residue in honey and wax. During treatment, the slow and continuous release of the active ingredient of this product causes a strip to affect various stages of the parasite's life cycle. These properties, along with the ease of use of this product, have made Flumvar a very suitable product in the beekeeping industry. This product has no side effects and does not cause death, reproduction or other disorders in bee colonies.

#### • Apiguard

The active ingredient of this compound is thymol and its amount is 5.12 grams per tray. Apiguard is a gel form of thymol and is easy to use. Each apiguard tray contains 50 grams of gel. This compound should be used in the fall after the baby has finished growing. In an experiment conducted in Italy, it showed up to 98% of Varoa deaths. But in a Canadian experiment, May showed a mortality rate of 82-88 percent. However, this test, which was performed in Canada, was not performed in the fall as suggested. (5)

#### Non-chemical methods

Here are some non-chemical ways to fight Varroa:

#### Using heat

Mite Varroa is unable to survive on adult bees at 47. C. In this method, the beekeeper must take all the insects out of the infected hive and put them in a place for 10 minutes at a temperature of 47 degrees. In this method, 95% of the mites are destroyed (9).

#### Absorbent traps for Varroa

In this method, researchers have tried to kill females by inserting a female varroa into traps containing bees (preferably male bees) and pheromones released from the baby, and the varroa are interested in it (5).

Sprinkling small rounds on bees Ramirez tried to tell Varroa from his host by dipping the bees' powder with different types of powders (glucose, flour, powdered salt, etc.) in order to disable the female Varroa balloons with which he clings to the host. The method has arrived. In the glucose laboratory, it was useful in all samples a few hours after use, other powders were less effective than glucose, and the overall success rate was between 87-97%. But the disadvantage of this method is that the bee, which is full of dust and dust, starts to clean itself after a short time. With this method, Varroa may leave the host for a short time, but these bees do not seem to secure their body system (5).

#### Hybridization and breeding

In this method, hybrids are obtained from two types of bees, one of which is more resistant to varroa, and thus a defensive state is created against varroa mite (5).

#### **Anti-Varroa frames**

In this way, the growth and development of workers and males is shortened, which in turn shortens the time Varroa needs to reproduce. Beekeepers replace ordinary hive frames with frames that have large rooms. The queen always lays female eggs in multiple small cells and male eggs in relatively large cells in a normal frame, but when only large cells are available, the queen's behavior changes, leaving most female eggs in anti-Varroa frames. As worker bees fill these cells with more food, the maturation of female larvae and nymphs accelerates. Those who use these frames believe that this maturity between 17-19 days instead of 21 days of normal conditions will prevent the re-production of Varroa (5).

#### **Biological methods**

• Use of male cell-free frames: If there are no male cells, the mites invade the worker cells, and since the worker bee cycle is not enough, the mites are less likely to be successful in spawning (1).

• Use of new frames: Black frames that have been used for a long time have narrower cells and the size of the born bees will be smaller and the power of these bees to fight against mites as well as honey production will be reduced (7).

• Use of male cell frames: Given that Maro Varroa is highly dependent on bee infants to complete its life cycle and prefers male infants, this weakness of mite can be used to control it. For this purpose, the male infants can be placed inside the hive and given to the queen to lay eggs and raise the baby. Mite Varroa quickly enters these cells and concentrates on the male larvae. After the cells of these shanks are closed, we take them out of the hive and destroy them (6).

#### **Imprisonment of the Queen**

In this method, in order to prevent the queen from laying eggs, the queen is locked in an empty frame for 7 days. The frames remain in the hive until the door of the room is closed, after which they are removed from the hive and destroyed. Experiments have shown that in this method, an average of 80% (sometimes up to 98%) of mites in the hive is lost (10).

## Sterilization of male and female mites

In this method, using x-rays or other ionizing radiation, male and female mites are sterilized and mite reproduction is stopped (1).

#### Microbiological struggle

This method uses microorganisms that are not dangerous to bees but kill mites (1).

**Materials and equipment required** To perform the research steps, the following materials and equipment are required, which were prepared according to the plan.

Expanded glass door, large plastic funnel, detergent liquid, sieve, pliers, reading lamp, cream collecting needle, paper tape, waterproof magic, magnifying glass, magnifying glass, fluorescent tape, beekeeping clothing (gan).

#### Selecting beekeeping and bee colonies

First, at the end of the summer of 1392, a beekeeping was selected in the Shemiranat area of Tehran, Iran. Bees and 3 baby frames were selected. The hives were not treated for Varroasis veins in the year under review.

Group 1 (c): As a control group (control) do not include treatment.

Group 2 (F): As a group treated with flumvar Group 3 (b): As a group treated with bayvarol

**Determination of the initial contamination of bee colonies to varroa** After all the hives in each group have been numbered with a pen, remove one of the hives from each hive and insert the bees onto it through a funnel and brush into the wide-mouthed glass so that about 200 adult bees enter each glass. Each glass was coded with paper tape and transported to the laboratory.

At the same time, one of the hives containing unopened pupae was removed from each hive, separated by 200 cells, and placed in a nylon bag. After coding, it was transferred to a laboratory and stored in the refrigerator until it was examined.

## Separation of Varroa mite from adult bees

#### Assiut Veterinary Medical Journal

To separate the Varroa mite from adult bees, some lukewarm water was added to the glass containing the bee, then a few drops of dishwashing liquid were added, and after closing the glass door, it was shaken a little and left for an hour. Then, after shaking the contents of each glass for 10 seconds on a surface on which the sieves were emptied and the bees remained on top of the sieve net, the Varroa mites were separated and taken to the bottom and counted in the next step. In addition, the isolated bees were studied one by one under the light of the study lamp and with the help of a magnifying glass, so that if there was any undifferentiated mite left on them, it would be separated and counted. Of course, most of the mites were separated from the bees and counted.



Figure 2. Steps to isolate whiteflies from adult bees

## Separation of mites from bee babies in the chamber of pupae

In order to separate the mites inside the pupal cells, the door of each cell was slightly opened with the help of a worm-collecting needle, and the baby bee was taken out of it, and the possible mites were separated from them and counted. Also, inside each cell, under the light of the study lamp, the remaining mites inside the cells were separated and counted.

In the end, the number of isolated mites in all three groups was recorded in special forms. From each hive, 200 chambers of pupae were examined and examined.



Figure 3 Steps to separate Mite Varroa from pupae

## Using drugs in the target groups of beehives

In each hive of Group 2 (F), a flumer strip was placed between boxes 3 and 4

according to the manufacturer's instructions.

## Determining the amount of infection of pupae of each group of hives 40 days after taking the drug

Forty days after taking the drug from each hive, another 200 pupae were examined and counted.

## Determining the level of contamination of bee hives in each group 40 days after taking the drug

After 40 days, as in the initial examination of bee contamination with varroa, the amount of bee contamination was investigated and the results were recorded.

## Statistical analysis of the results After performing all the previous steps, the results

It was reviewed and analyzed by SPSS software. For this purpose, the subject of differences between groups was first investigated by ANOVA test (analysis of variance). Explaining that in order to ensure the continued success of the project, after determining the initial level of infection of pupae and bees of all groups, ANOVA test was performed before taking the drug to ensure that all groups were infected with MIT. Varua did not differ significantly from each other, which fortunately the results of the initial study indicated that there was no significant difference, and therefore the degree of confidence in the results of the study was scientifically and statistically acceptable. The rate of infection between pupae was compared with T-test before and 40 days after treatment. This was done by comparing the amount of primary infection and then treatment in adult bees. Multiple comparisons between groups were also performed using the Dennett C posttest method.

## RESULTS

According to subsequent studies, the rate of varroa contamination decreased from 12% to 2%, respectively, following the use of bivarium and fluorine. In the negative control group, the rate of contamination increased from 12 to 15% due to non-use of the drug. The therapeutic effect should be determined by bivalve and fluoride as 82.54 and 90.26%, respectively, and the results are presented in Tables 1 and 2.

# **Table 1:** Comparison of contamination of Varroa-infected hives following flumavar and bayvarol treatment in a number of female beekeepers in Tehran province

	Group		Experimental (flumavar)	
	Control (negative) before the treatment	Control (negative) after the treatment	Before flumavar treatment	After flumavar treatment
	13	17	15	2
First	11	14	13	1
Repetition	9	13	11	1
	13	18	14	2
	16	20	10	1
	10	13	10	1
Second	10	14	11	1
Repetition	12	15	13	2
	14	18	11	2
	11	14	9	1
	12	16	12	1
Third	9	14	10	0
Repetition	14	13	9	1
	15	15	13	1
	10	12	13	0

**Table 2:** Comparison of the therapeutic effect with biovarul and flumavar

	Efficacy (%)	Standard Error	Standard Devicrtion
flumavar	90.26b	± 0.115	± 0.029
Descriptions: A and B indicate a significant difference between the two numbers. + (p <0.05)			

## Examining the possible side effects of medication

Due to the continuous control and visit of the hives during the period of 40 days after the use of bivalve and fluorine, no possible complications were observed in adult bees and pupae.



Figure 1: Comparison of adult bee infection with varroa in three groups (before treatment)



Figure 2: Comparison of adult bee contamination with varroa in three groups (after treatment)

As can be seen in Tables 1 and 2, the fluoride has reduced the initial contamination rate of varroa from 11% to 1%, resulting in a therapeutic effect of 90.26% (P <0.05) Also, according to the controls, no side effects were observed due to the use of both drugs.

Flum-like has been used with the active ingredient Flumtrin in other countries. In a Uruguayan study, fluoride contamination was reduced by 70 percent in 24 hours following flumers. According to the study, the fluoride efficiency increases to more than 90% after one week (12).

Due to the fact that the phloem remains in the hive for thirty days, the stability of its therapeutic effect causes the bees that come out of the pupae and are possibly infected to be killed by the venom. In another study in three geographical regions in Argentina conducted by Del Hoyo *et al.*, The rate of fluoride-like therapeutic efficacy was 98.80, 97.39 and 98.9%. In this study, different climatic conditions did not affect the fluoride efficiency.

In another study conducted in 2008-2007 in Slovenia by Smodis Skerl et al. The efficiency of flumerine and oxalic acid has been compared with each other. In this study, flumetrin and oxalic acid reduced the 24% and 12% of the Varroa population, respectively. (17).

Another study in the Bursa region of Turkey in 2019 looked at the therapeutic effect of flumetrin after a 4-week course of treatment. Its therapeutic effect is reported to be 94%. (18). In another study conducted in 2019 by Ferrer and colleagues. The effect of flumerine antiviral effect with fluvolatin has been studied. After a 28-day treatment period, both toxins had a therapeutic effect of over 95%. (15).

In another study, between 2003 and 2015, October 2 and December were performed using 5 aparicidan acaricides, with bayvarol, Apivar, Prrizin and Flumtrin. The highest therapeutic effect with 96% was related to Flumtrin. And the epistle to 85% had the least therapeutic effect. 95% of opiates, 89% of bayvarol and 94% of prismine had anti-Varroasis treatment efficacy. (16).

The reduction in the therapeutic effect of apistane by 85% and bayvarol by 89% are attributed to the phenomenon of drug resistance.

In another study conducted in Slovenia in 2016, fluorine and fluvalinate were still compared. Both poisons have been able to significantly reduce contamination, with a reported therapeutic effect of 96.24% and 94.3%, respectively.

In their study in Slovenia, George and Scroll (2007) examined the therapeutic effects of flumerine and fluvalinate, both of which accounted for more than 90%, but the therapeutic effects of fluvolatin in some areas were slightly higher than flumerine, some of which differed. Significantly reported at the level (P <0.05). (17).

Scarlett *et al.* (2011) again compared the effect of flumetrine with oxalic acid in Slovenia in some areas, the effect of fluoride has been greater, and in some areas, the therapeutic effect of oxalic acid has been greater. (17).

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