

Pettis Test - Detecting Varroa Mite Resistance to Apistan, Apivar & Coumaphos

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The information on this factsheet has been modified with permission from Dr. J. Pettis of USDA-ARS Bee Research Laboratory, Beltsville, Maryland.

This test can be used to determine mite resistance when a beehive doesn't appear to respond to chemical mite control measures. It uses fluvalinate (Apistan®), amitraz (Apivar) or coumaphos (CheckMite+TM) strips and can test all three products simultaneously on the same group of colonies by taking three samples from each hive. Each sample, whether Apistan, Apivar or CheckMite, requires one jar.

Materials required for each test:

- 500ml jar with lid (wide-mouth canning jar)
- light metal mesh cover for the jar (8 mesh to the inch hardware cloth)
- index card (or similar) and stapler
- 3/8" X 1" piece of a new Apistan® or CheckMite+TM strip
- 1/4cup (60 ml) measure to scoop up bees
- 25% alcohol or windshield washing fluid
- straining cloth and 6 clothes pins
- plastic or rubber gloves
- plastic bucket
- sheet of white paper



Step 1

Staple a 1 cm x 2.5 cm (3/8" X 1") section of an Apistan®, Apivar or CheckMite+TM strip to the center of an index card. Wear gloves when handling these strips. Place the card in a jar with the miticide section facing inwards. Replace the solid, round metal section of the canning jar lid with a piece of wire mesh. The holes in the mesh should be large enough to let Varroa through easily.

Step 2

Select one or two brood frames and make sure there is no queen on the comb. Shake bees from the frame(s) onto an up-turned hive lid, or into a bucket or box. Scoop up 1/4 cup or 60 ml of bees (~150 bees) and place them into each jar, being careful not to damage the bees. Screw the lids on the jars to stop the bees from escaping.

Step 3

Place the jars in an incubator or a warm room, in the dark, for 6 hours. Alternatively, place jars in a picnic cooler with a couple of hot water bottles. Refill the bottles with hot water after the first 3 hours. Make sure the lids of the jars are not covered so that the bees have air.

Step 4

After 6 hours, hold the jar about 10cm above a piece of white paper and turn upside down so that the mesh lid is facing downwards. Hit the jar with the palm of your hand three times. Count the number of mites that fall on the paper.

Step 5

Knock the bees to the bottom of the jar. Remove the index card with the attached strip and fill the jar half-way with alcohol or

washer fluid. This should be done outside, using gloves. Remove the mesh lid and replace with the original solid lid for the jar. Shake the jar vigorously for 5 min.

Step 6

Remove the solid lid and replace it with the mesh lid. Pour the fluid into the straining cloth pinned to the bucket. Refill the jar with fluid, swirl the bees around and pour through the strainer again.

Step 7

Count the number of mites recovered on the cloth. If the total number of mites recovered in each of the samplings is less than 5, the results should be discarded.



Step 8

To calculate the percentage of mites killed by Apistan®, Apivar or CheckMite+™, divide the number of mites that initially fell on the white paper before the bees were killed, by the total number of mites killed (*Note that the total mites killed = white paper mite count + mite count from bee washing*).

% kill by Apistan®, Apivar or CheckMite+™ = initial kill, divided by total mites x 100

If more than 50% of the mites were killed by the Apistan®, Apivar or CheckMite+™ after 6 hours, the mites are susceptible and adequate mite control can be expected. **If less than 50% of the mites are killed after 6 hours by Apistan®, Apivar or CheckMite+™, the mites are resistant.**

Critical Factors for the Success of the Resistance Test

- Prescreen hives using the ether roll technique (250-300 bees) and test only hives yielding 5 or more mites. This test gives meaningful results only when performed on hives with adequate mite levels. Do not expect levels of resistance to be the same among hives. Select 12 hives per apiary. More hives are better. This test is not designed to identify individual hives showing resistance. Use apiary averages to assess the results.
- Perform the test exactly as described. Jar size, size of Apistan® or CheckMite+™ pieces and temperature are important.
- Ensure that bees are mobile in the jars so they contact the strips. **Cool temperatures may cause the bees to cluster away from the strips.** If using darkened incubator, it may be helpful to open the incubator periodically to admit light and fresh air to encourage bee movement.
- It is best not to reuse strip pieces or index cards. Wash jars between tests.

Note: Do not expose jars with miticide sections to sunlight for any length of time. It is best to keep the jars in their storage boxes before and after filling until they are incubated. Sample the bees from brood frames. For accuracy and to avoid bee injury, use a measuring scoop. Do not scrape bees directly into jars.

Disclaimer: This assay is intended to screen for resistant mites and is not intended to indicate the exact level of resistance.