



## Apistan® (fluvalinate) and CheckMite+™ (coumaphos) Resistance Testing

### Are you using the right miticide? Are your mites resistant to Apistan® or CheckMite+™? Use this test BEFORE treating your bees.

This test can be used to determine mite resistance when a beehive doesn't appear to respond to chemical mite control measures. The test uses Apistan® and CheckMite+™ strips and can be conducted simultaneously on the same group of colonies by taking two samples from each hive.

Materials required to carry out the test:

- 500ml jar with lid (wide-mouth pint canning jar)
- light metal mesh cover for the jar (8mesh to the inch hardware cloth)
- index card or similar and stapler
- 3/8" X 1" piece of a new Apistan® and CheckMite+™ strip
- 1/4cup or 2oz. 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 3/8" X 1" section of an Apistan® and CheckMite+™ strip to the center of an index card. Make sure to handle the Apistan® and CheckMite+™ with gloves. Place the card in the jar with a section of the Apistan® or CheckMite+™ strip 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 easily let varroa through.

### Step 2

Shake bees from one or two brood combs into an up-turned hive lid, bucket or box. Scoop up 1/4 cup or 2oz. of bees (~150 bees) and place them into each jar, being careful not to damage the bees. Screw the lids onto the jars to stop the bees from escaping.

### Step 3

Place the jars in an incubator or a warm 30° C room in the dark for 6 hours. Make sure the lids are not covered so air gets to the bees.

## Step 4

After 6 hours, hold the jar about 10cm above the piece of white paper and turn it so 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 the 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 both samplings (Apistan®, CheckMite+™) is less than 5, the results should be discarded.

## Step 8

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

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



**Resistance test with bees**

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

## Critical Factors for the Success of the Resistance Test

- Prescreen hives using the ether roll technique (250-300 bees) and test only those 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 Apistan® or CheckMite™ 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 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.**

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