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P R O E X A G
NON-TRADITIONAL AGRICULTURAL EXPORT SUPPORT PROJECT

PROCEDURES MANUAL FOR MELON VIRUS EPIDEMIOLOGY IN CENTRAL AMERICA

Assignment Number: ST/90-04B

SUBMITTED TO:

Regional Office for Central America and Panama (ROCAP)
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Guatemala City, Guatemala

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April, 1990

PROCEDURES MANUAL FOR MELON VIRUS EPIDEMIOLOGY IN CENTRAL AMERICA

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OBJECTIVES:

1. Identify the important viruses present in each growing area
2. Identify the population densities of vectors throughout the growing season in each area
3. Identify the important vectors
4. When possible identify the major sources of vectors

GENERAL PROCEDURE:

Collection sites will be established at 10 to 15 field locations in each country. An insect trap will be set up at the northwest and southeast corner of each field. Once a week for the growing season at every trap site, aphids and whiteflies will be collected from the yellow pan traps, the level of virus infection will be estimated from 100 plants examined near the insect traps, and up to 10 samples from infected cucurbit plants will be collected. Insects will be preserved in alcohol, and plant tissue must be preserved frozen. Insects and plant tissue will be collected and stored in the country of origin and then transported to California for identification and assay.

SPECIFIC PROCEDURES:

FIELD DESCRIPTION:

Draw a map of each field at the beginning of the study which should include the location of insect traps, an estimate of the size of the field, compass directions and type of vegetation present on each side of the field for about a 30 meter distance. Examples of vegetation might be sugarcane, melons, trees, or weeds. Sometimes no vegetation is present and a disced field, river or habitations are present. Record this as well. Identify each trap by a field name and direction, for example: in field named Gomez, the two traps in the field might be Gomez-N and Gomez-S. Record an estimate of the date of planting, date of beginning harvest, the type of melon planted and cultivar if known. A sample form for this is included with this manual.

VIRUS INCIDENCE:

Evaluations of the incidence of virus-infected plants will be made in each field on a weekly basis by cooperators. One hundred plants will be chosen randomly at each trap site (a total of 200 plants per field) by walking away from the trap into the field and back again in a big loop of 30 to 50 m. diameter. As you walk through the field, examine each plant that your left foot lands beside. Keep a record as you walk whether the plant has virus symptoms or not. Examine a total of 100 plants. The number of symptomatic plants out of these 100 will be recorded as an estimate of the virus percentage in the field. Observe the the average growth stage of plants in the field at the same time and record it.

There are a number of different symptoms that can be caused by viruses. Mosaic, mottle, leaf distortion and leaf deformation are probably the most common symptoms. Others are stunted plants, chlorotic spots, and leaf curling. Many of these symptoms can be caused by other agents, a common one being the phytotoxic effects of some pesticides. When in doubt if a symptom is caused by a virus or some other cause, in almost every case, an infected plant will show symptoms in the youngest leaves.

COLLECTION OF PLANT SAMPLES:

As you are examining plants for virus incidence, collect a growing tip (apical tip plus youngest two leaves) of the first ten plants which have virus symptoms at each trap site. Use the plastic bags provided to collect and store the plant sample. Each bag should contain one plant sample and have the date, field-trap identification and a number (1 through 10) written on the bag with the waterproof marker provided. Put the 10 bags together into one bag, storing all samples of a given week and site together. Be sure to seal the bag well before placing it on ice, since water in the bag will complicate analysis. It is important that these samples be placed on ice within minutes after they are collected and kept cold until they are moved to a freezer for longer storage. Plant samples should be frozen within 24 hours of collection. Do not store plant samples for more than 12 hours on ice or in a refrigerator, and do not leave samples at room temperature. Once frozen, plant samples should never be thawed. UC-Riverside researchers will carry these plant samples on dry ice back to Riverside for later analysis. A supply of dry ice will need to be secured in advance by in-country cooperators.

APHID AND WHITEFLY TRAPPING:

At each field site, two plastic water pan traps will be placed in opposite corners of the field. A 15x15 cm yellow plastic card should be placed in the bottom of the trap. This card will discolor after about a week. Turn the card over and use the other side for the next week and then discard it and use a new one.

Fill the traps with a 50% mixture of ethylene glycol and water which contains a couple of drops of liquid dishwashing detergent. Use clear ethylene glycol (not colored as in antifreeze) or propylene glycol. One or the other should be locally available. The ethylene gly-

col retards evaporation and the detergent breaks the surface tension and permits insects to sink to the bottom of the trap. Use clean water to dilute the ethylene glycol, water from the irrigation ditches contains algae that catch on the aphids and prevent species identification. Trap fluid can be recycled by bringing the strained fluid back to the lab, letting it stand to settle out the fine dirt, pouring off the fluid to separate it from the dirt and adding water to the proper 50:50 consistency.

Once a week, collect insects from the traps by straining the insects from the trap fluid using the supplied funnels, strainer and wash bottles. Pour contents of trap through strainer collecting liquid in a container (save for recycling). Use a squeeze bottle containing water to rinse the trap, pouring rinse fluid through strainer. Rinse contents in the strainer with water to remove dirt. Turn strainer upside down over the collection vial and wash contents into vial with 70% ethanol in squeeze bottle. Remove strainer and replace with cap. Insects should be preserved in these plastic vials containing 70% ethanol (methanol does not work very well.) It is recommended that labels for insect vials be made before going out to the fields. Use pencil to write the date and field-trap identification on small strips of white paper which should be placed in vial after insects are collected. Store the vials in an upright position to minimize fluid loss.

Vial contents should be examined using a stereoscope and all insects except whiteflies and aphids should be discarded. Whiteflies should be counted, the number recorded on the proper form and then discarded. Aphid samples should be saved. Aphid samples will be collected by UC-Riverside researchers periodically and carried to Riverside for identification.

DESCRIPTION OF FIELD STUDIED

AREA:

FIELD:

CULTIVAR:

DATE PLANTED:

DATE BEG. HARVEST:

APPROXIMATE SIZE:

DESCRIPTION OF FIELD:

SURROUNDING AREAS:

1.	7.
2.	8.
3.	9.
4.	10.
5.	11.
6.	12.

1989/90

DESCRIPTION OF FIELD STUDIED

AREA: 25 MZ

FIELD: El Cocal

AN EXAMPLE

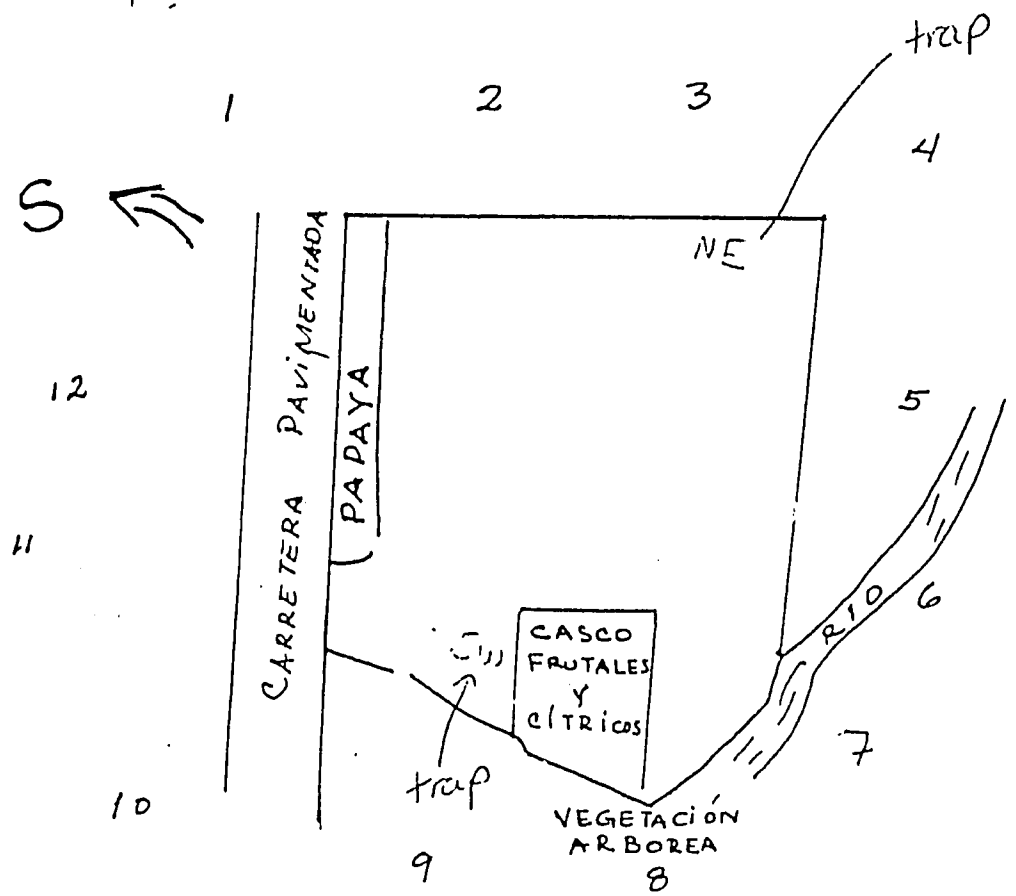
CULTIVAR: melon (canicloupe)

DATE PLANTED:

DATE BEG. HARVEST:

APPROXIMATE SIZE: -

DESCRIPTION OF FIELD:
(Mapa)



SURROUNDING AREAS:

1. Caña de azucar
2. Pasto
3. Pasto
4. Pasto
5. Zacate de corte
6. Zacate de corte
7. } Vegetación
8. } arbustiva
9. Musáceas (plátanos)
10. }
11. } pequeña área Sembrada
12. } con papaya.

SUMMARY OF VIRUS VECTOR FIELD DATA

FIELD SITE:

DATE	NO. APHIDS	NO. WHITEFLIES	PERCENT VIRUS INF.		

6'

FIELD HOST DATA

FIELD SITE:

DATE	MEAN NO. OF LEAF/BRANCH	MEAN SIZE OF FRUIT	NO. SAMPLES COLLECTED	OBSERVATIONS