

Technique for determination of host status of fruit flies (Tephritidae)



Suputa

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Referenses

ISPM 37

- <https://www.ippc.int/en/core-activities/standards-setting/ispms/>
- https://www.ippc.int/static/media/files/publications/en/2015/01/26/cpm_2015_06_01_2006-031_draftispm_ffhoststatus_2014-12-24.pdf

The Guideline of Fruit Fly (Diptera: Tephritidae) Collection and Preservation.

Suputa, Cahyaniati, A.T. Arminudin, A. Kustaryati, M. Railan, & Issusilaningtyas, 2007. Ministry of Agriculture. Indonesia. ISBN 978-979-3147-28-4

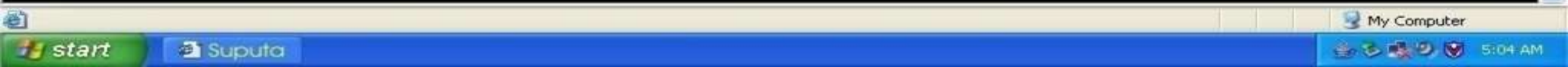
Determination of host status of fruit to fruit flies

Fruit as referred to in this standard covers fruit in the botanical sense, including such fruits that are sometimes called vegetables



Host Status

Classification of a plant species or cultivar as being a natural host, semi-natural host or nonhost for a fruit fly species



Natural Host

A plant species or cultivar **that has been** scientifically **found to be** infested by **the** target fruit fly species **under** natural conditions **and able** to sustain **its** **development** to viable adults



Semi-Natural Host

A plant species or cultivar **that** is not a natural host **but has been** scientifically demonstrated to be infested **by the** target fruit fly species **and able** to sustain **its development** to viable adults **as concluded from** the semi-natural field conditions set out in this standard

Non-Host

A plant species or cultivar **that has** not been found to be infested **by the** target fruit fly species **or is not able** to sustain **its development** to viable adults **under natural conditions or under the seminatural field conditions set out in this standard**

It is important to note that host status **may change** over time because of changes in biological conditions

Historical records and published reports may sometimes be unreliable, for example:

- Fruit fly species and plant species or cultivars **may have been** incorrectly identified **and** reference specimens **may not be** available **for verification.**

Historical records and published reports may sometimes be unreliable, for example:

- Important details may have been omitted (e.g. cultivar, stage of maturity, physical condition of fruit at the time of collection, sanitary condition of the orchard).

A screenshot of a web browser window. The address bar shows 'http://hpt.faperta.ugm.ac.id/suputa/'. The browser interface includes a menu bar (File, Edit, View, Favorites, Tools, Help), a toolbar with icons for Back, Forward, Stop, Refresh, Home, Search, Favorites, Media, and other functions, and a taskbar at the bottom with a Start button, a 'Suputa' taskbar icon, and a system tray showing 'My Computer' and the time '5:04 AM'. The main content area is a black slide with yellow and white text.

Historical records and published reports may sometimes be unreliable, for example:

- Development of larvae **to viable adults may not have been** verified.

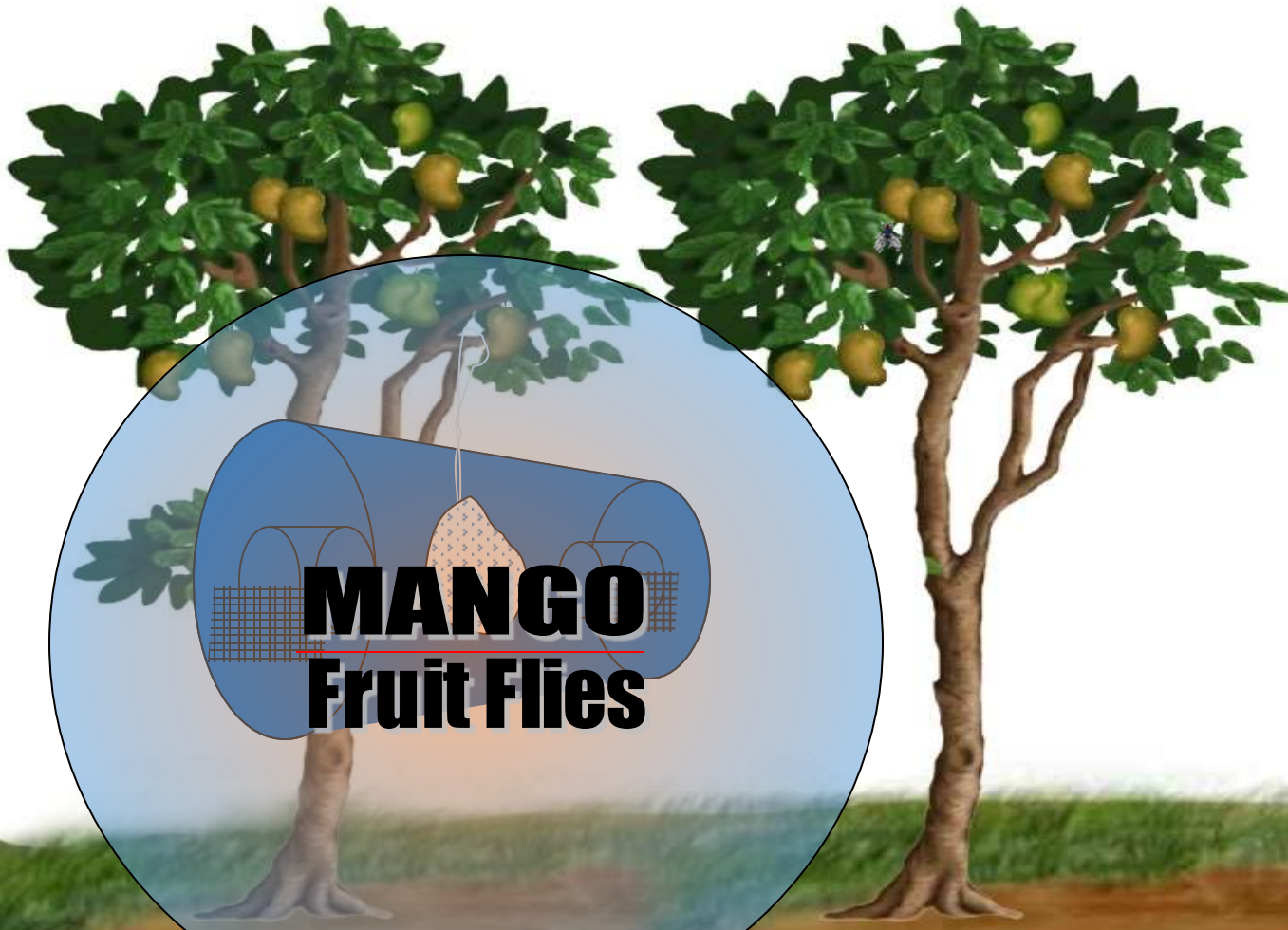
Historical records and published reports may sometimes be unreliable, for example:

- Collection records **may be** incorrect or dubious

Host status based on:

1. damaged fruit;
2. simply finding larvae inside fruit;
3. cross-contamination **of samples;**
4. **the** catch from a trap **placed on a fruit plant** (Common Habit).

Misinterpretation
TRAPPING
METHYL EUGENOL



MANGO
Fruit Flies

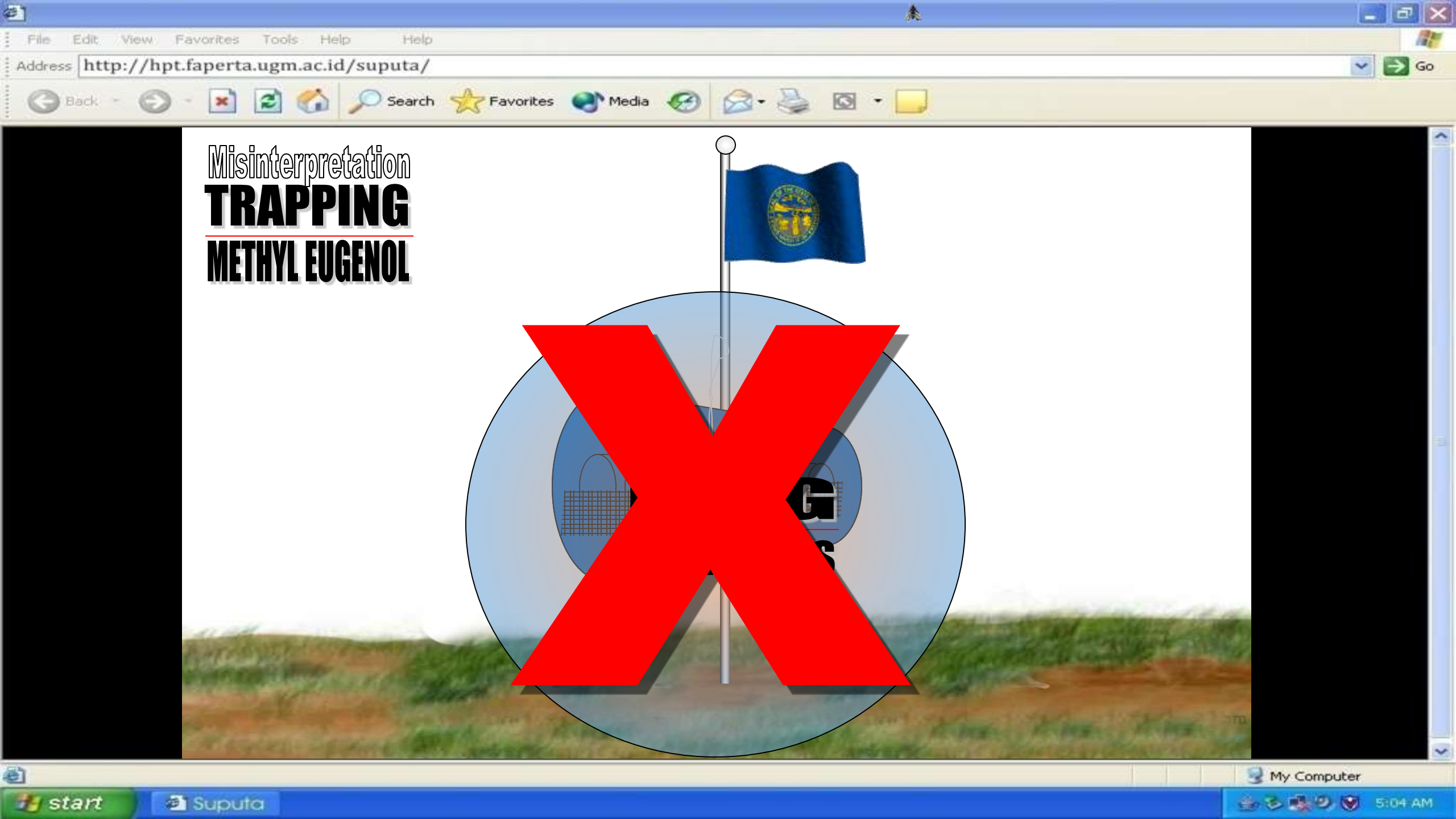
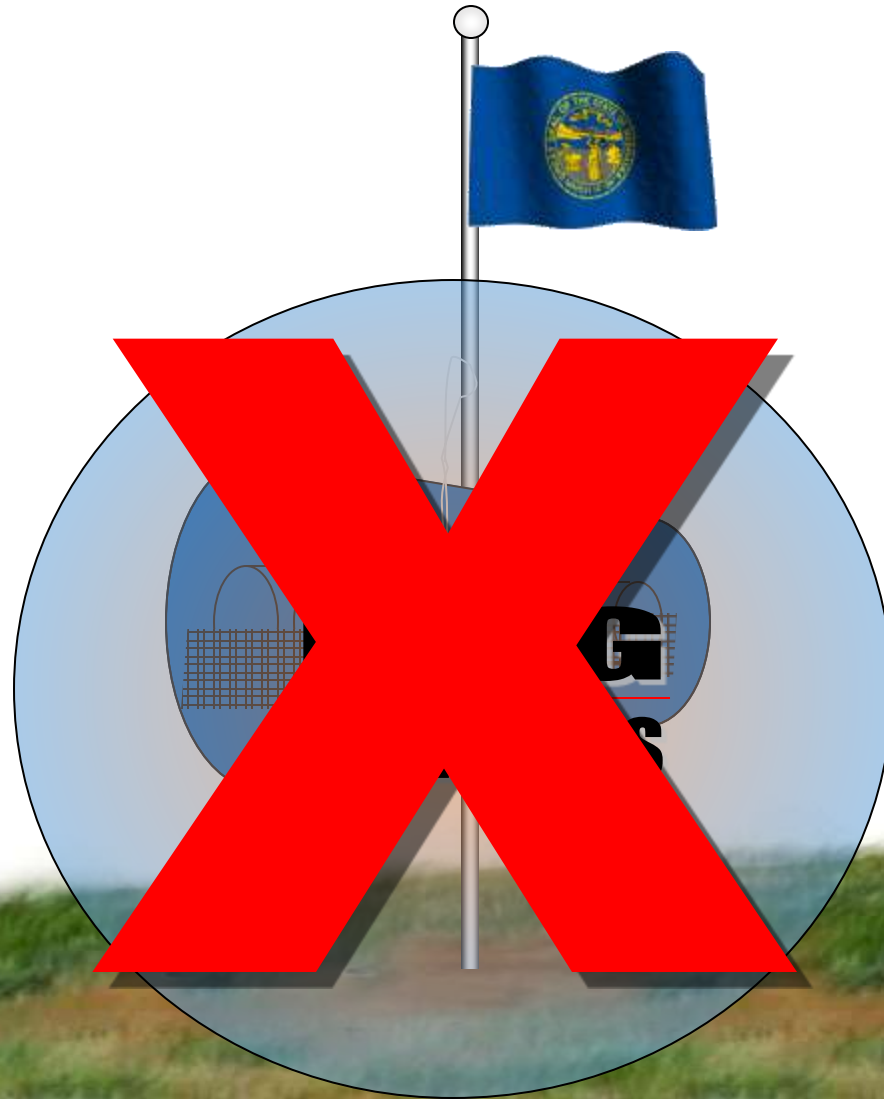
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Misinterpretation
TRAPPING
METHYL EUGENOL



Misinterpretation
TRAPPING
METHYL EUGENOL

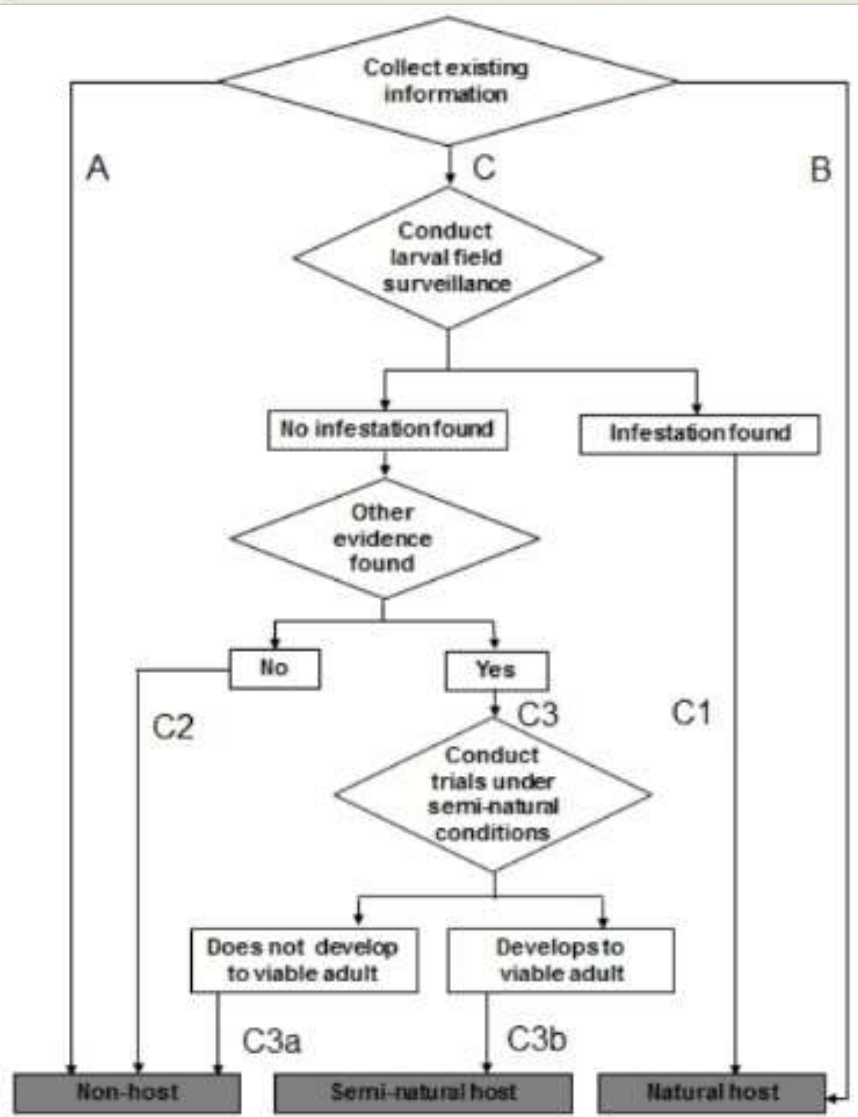


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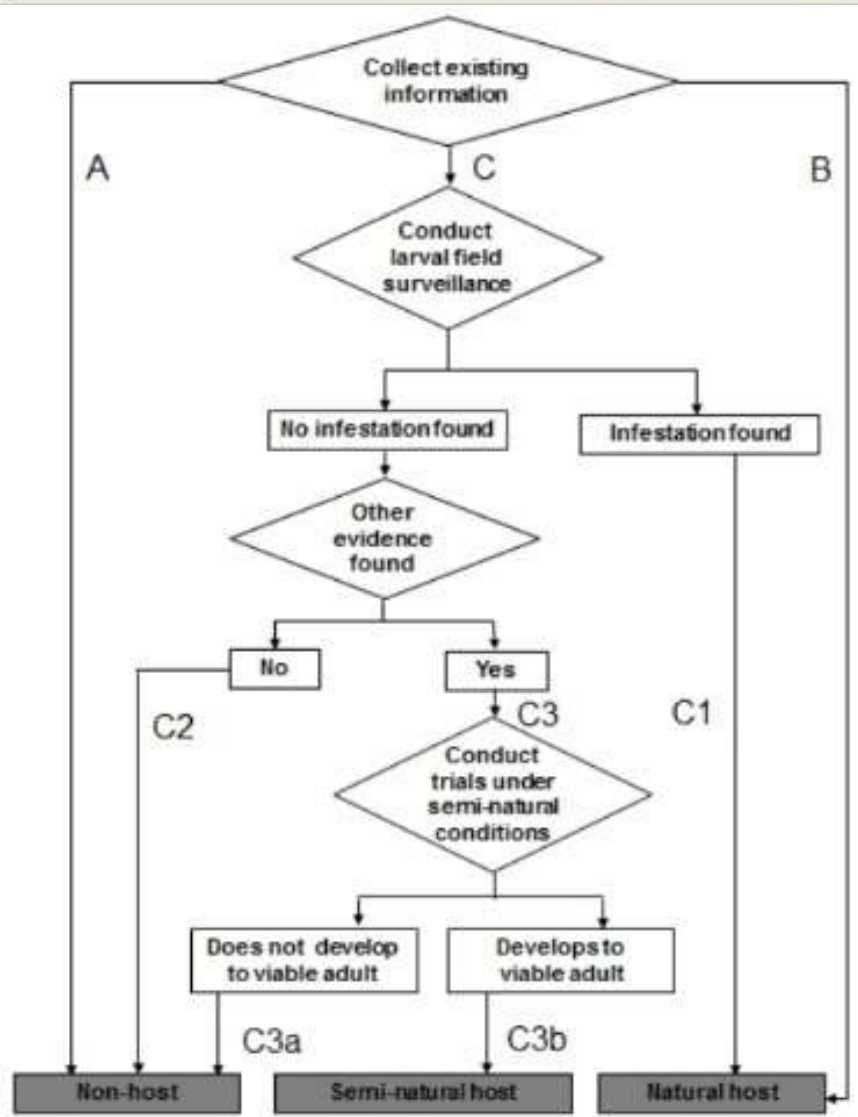
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Steps for the determination of host status of fruit to fruit flies

- A.** When existing biological or historical information provides sufficient evidence that the fruit does not support infestation and development to viable adults, no further surveys or field trials should be required and the plant should be categorized as a **non-host**.
- B.** When existing biological and historical information provides sufficient evidence that the fruit supports infestation and development to viable adults, no further surveys or field trials should be required and the plant should be categorized as a **natural host**.



Steps for the determination of host status of fruit to fruit flies

C. When existing biological and historical information is inconclusive, appropriate field surveillance by fruit sampling or field trials should be used to determine host status. Surveillance and trials may lead to one of the following results:

C1. If infestation with development to **viable adults** is found after **field surveillance by fruit sampling**, the plant should be categorized as a **natural host**.

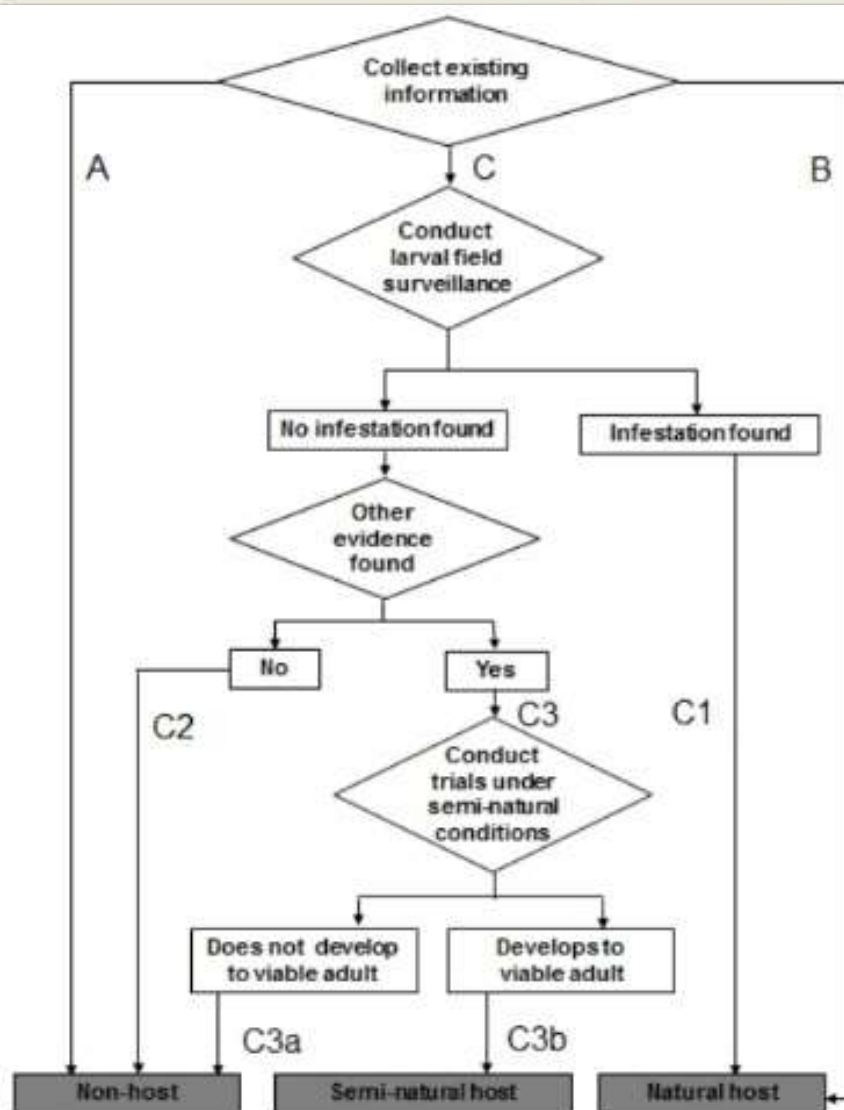
C2. If **no infestation** is found after field surveillance by fruit sampling, and no further information indicates that the fruit has the potential to become infested, the plant may be categorized as a **non-host**.

Steps for the determination of host status of fruit to fruit flies

C3. If no infestation is found after field surveillance by fruit sampling, but available biological or historical information indicates that the fruit has the potential to become infested, additional field trials under seminatural conditions may be needed to assess whether the target fruit fly can develop to viable adults on the particular fruit species or cultivar.

C3a. If the target fruit fly species does not develop to viable adults, the plant should be categorized as a non-host.

C3b. If the target fruit fly species does develop to viable adults, the plant should be categorized as a seminatural host.



The following elements are important considerations in planning field trials:

- the identity of the plant species (including cultivars where appropriate) and the target fruit fly species
- the physical and physiological variability of the fruit in the production area
- past chemical usage in the fruit production area

The following elements are important considerations in planning field trials:

- target fruit fly incidence over the entire production area, and relevant harvest and export periods
- relevant information, including literature and records, regarding host status of the fruit and fruit fly species, and a critical review of such information
- the origin and rearing status of the fruit fly colony to be used

The following elements are important considerations in planning field trials:

- **known** natural host species and cultivars **to be used as** controls
- separate field trials, **where appropriate, for** each fruit fly species **for which determination of host status is required**
- separate field trials **for** each cultivar **of the fruit if cultivar differences are the purported source of host variability to infestation**

The following elements are important considerations in planning field trials:

- the placing of field trials in the fruit production areas
- compliance with sound statistical practice.

1. Natural Host Status Determination Using Surveillance by Fruit Sampling

- Fruit sampling **is the most** reliable method to determine natural host status.
- Fruit samples **should be** representative of the range of production areas and environmental conditions, as well as of physiological and physical stages.

2. Host Status Determination Using Field Trials under Semi-natural Conditions

- The objective of field trials is to determine host status under specified conditions of a fruit that has been determined not to be a natural host. Trials may include the use of field cages, greenhouses (including glass, plastic and screen houses) and bagged fruit-bearing branches.
- The emergence of a viable adult in any one replicate of a field trial under semi-natural conditions indicates that the fruit is a conditional host.
- The following subsections outline elements that should be taken into account when designing field trials

2.1 Fruit sampling

The following requirements apply to fruit sampling in field trials:

- Where possible, sampling should target fruit suspected of being infested.
- Period of time, the number of repetitions per growing season and the number of replicates should account for the variability of target fruit flies and fruit over time and over the production area. They should also account for early and late harvest conditions and be representative of the proposed area from where the fruit will be moved. The number and weight of the fruit required and replicates per trial to determine effectiveness, and appropriate confidence level, should be specified.

2.2 Fruit flies

The following requirements apply to operational procedures pertaining to the fruit flies used in field trials:

- Taxonomic identification of the fruit flies used for the field trials should be performed and voucher specimens be preserved.

2.2 Fruit flies

The following requirements apply to operational procedures pertaining to the fruit flies used in field trials:

- Basic information on target fruit fly species, including normal period of development and known hosts in the specific production area, should be compiled.

2.2 Fruit flies

The following requirements apply to operational procedures pertaining to the fruit flies used in field trials:

- The use of wild populations for the field trials is desirable. If wild flies cannot be obtained in sufficient numbers, the colony used should not be older than five generations at the initiation of the trials, whenever possible. The fruit fly population may be maintained on substrate, but the generation to be used in the trials should be reared on the natural host to ensure normal oviposition behaviour. Flies used in experimental replicates should all come from the same population and generation (i.e. cohort).

2.2 Fruit flies

The following requirements apply to operational procedures pertaining to the fruit flies used in field trials:

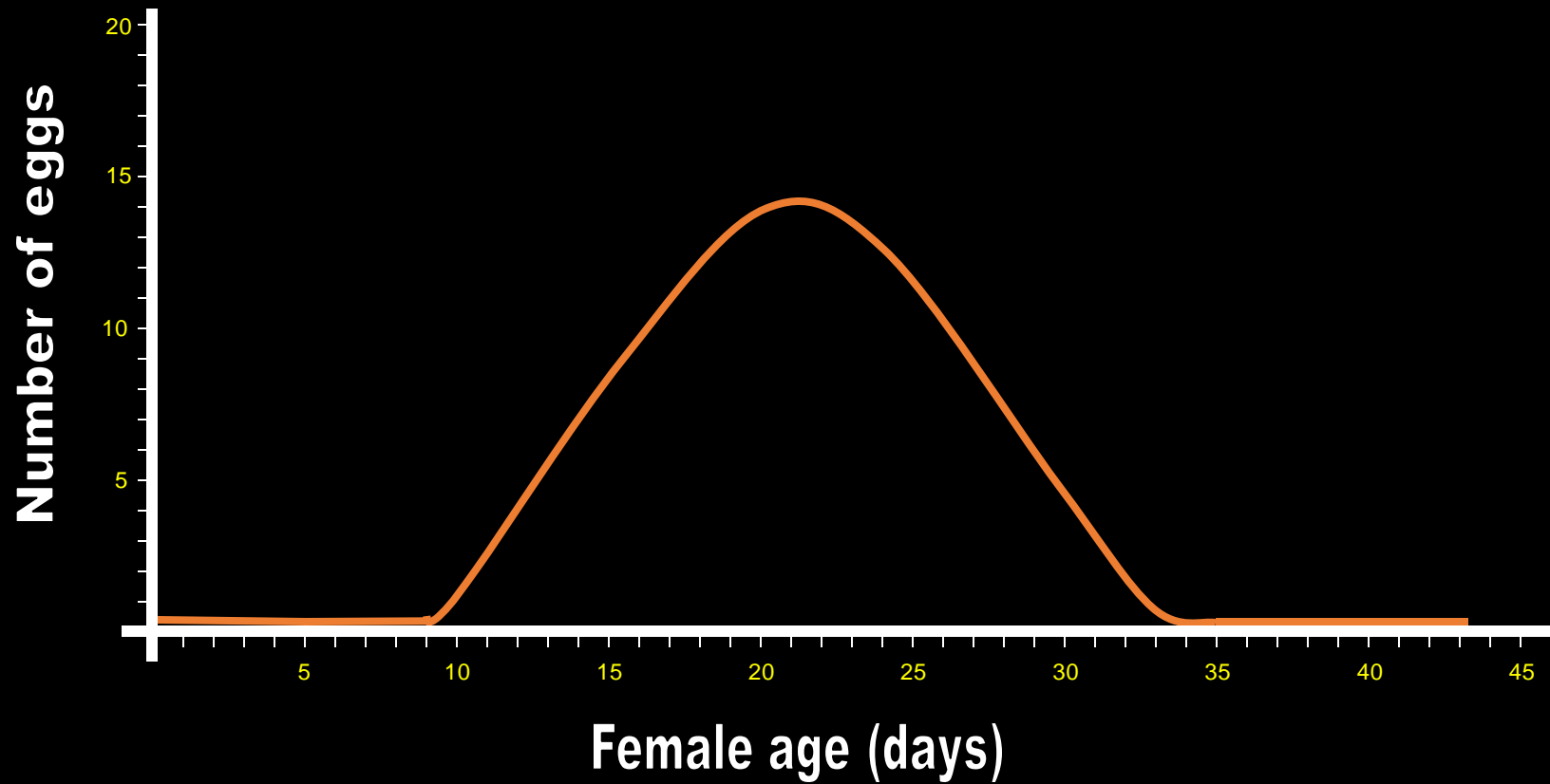
- The fruit fly colony should originate from the same area as the target fruit whenever possible.

2.2 Fruit flies

The following requirements apply to operational procedures pertaining to the fruit flies used in field trials:

- Pre-oviposition, oviposition and mating periods **should be determined before the field trials so that mated female flies are exposed to the fruit at the peak of their reproductive potential.**

OVIPOSITION



Suputa, 2007

2.2 Fruit flies

The following requirements apply to operational procedures pertaining to the fruit flies used in field trials:

- The age of the adult female and male flies should be recorded on the mating date and at the beginning of the field trials.

2.2 Fruit flies

The following requirements apply to operational procedures pertaining to the fruit flies used in field trials:

- The exposure time of the fruit to the target fruit fly species should be based on fruit fly oviposition behaviour.

2.2 Fruit flies

The following requirements apply to operational procedures pertaining to the fruit flies used in field trials:

- An individual female fly should be used only once.

2.2 Fruit flies

The following requirements apply to operational procedures pertaining to the fruit flies used in field trials:

- The number of adults dying during the field trials should be recorded and dead fruit flies should be replaced with live adults of the same population and generation (i.e. cohort). High adult mortality may indicate unfavourable conditions (e.g. excessive temperature) or contamination of field trial fruit (e.g. residual pesticides). In such cases, the trials should be repeated under more favourable conditions.

2.2 Fruit flies

The following requirements apply to operational procedures pertaining to the fruit flies used in field trials:

- The number of mated female flies required per fruit **should** be determined according to fruit size, female fecundity and field trial conditions. The number of fruit flies per replicate trial **should be determined according** to fruit fly biology, amount of fruit to be exposed and other field trial conditions.

NUMBER OF LARVAE PER FRUIT



Photo: SUPUTA
Gadjah Mada University

- Melon
- Rambutans
- Guava
- Gnetum
- Mango
- Carambola

Suputa, 2007

2.4 Controls

Fruit of known natural hosts at known stage of maturity are required as controls for all field trials. These may be of different species or genera from the target fruit species. Fruit should be free of prior infestation (e.g. by bagging or from a pest free area). Fruit flies used in controls and experimental replicates (including control) should all come from the same population and generation (i.e. cohort)

Controls are used to:

- verify that female flies are sexually mature, mated and exhibiting normal oviposition behaviour
- indicate the level of infestation that may occur in a natural host
- indicate the time frame for development to the adult stage under the field trial conditions in a natural host
- confirm that environmental conditions for infestation are appropriate.

2.5 Field trial design

Fruit flies are released into large mesh field cages that enclose whole fruit-bearing plants or mesh bags that enclose the parts of plants with the fruit. Alternatively, fruit-bearing plants may be placed in greenhouses into which flies are released.

It is important to note that because female fruit flies are artificially confined within the specific enclosure under observation, they may be forced to lay eggs in the fruit of a conditional host.

The female fruit flies be forced to lay their eggs in the plastic glass



2.5 Field trial design

Field trials should be conducted under conditions appropriate for fruit fly activity, especially oviposition, as follows:

- Field cages and greenhouses should be of an appropriate size and a design to ensure confinement of the adult flies and trial plants, allow adequate airflow and allow conditions that facilitate natural oviposition behaviour.
- Adults should be provided with satisfactory and sufficient food and water.
- Environmental conditions should be optimal and be recorded during the period of the field trials.
- Male flies may be kept in cages or greenhouses with the female flies if it is beneficial for encouraging oviposition.
- Natural enemies to the target fruit fly species should be removed from the cages before initiating the trials and re-entry should be prevented.
- Cages should be secured from other consumers of fruits (e.g. birds and monkeys).

2.5 Field trial design

- For controls, fruit from known natural hosts can be hung on branches of plants (not on the branches with test fruit). Controls must be separated from test fruits (in separate field cages, greenhouses or bagged fruit-bearing branches) to ensure the trial is not a choice test.
- The test fruit should remain naturally attached to plants and may be exposed to the fruit flies in field cages, bags or greenhouses.
- The plants should be grown under conditions that exclude as far as possible any interference from chemicals deleterious to fruit flies.
- A replicate should be a bag or cage, preferably on one plant at the experimental unit.
- Fruit fly mortality should be monitored and recorded and dead flies immediately replaced with live flies from the same population and generation (i.e. cohort) to maintain the same fruit fly incidence.
- The fruit should be grown under commercial conditions or in containers of a size that allows normal plant and fruit development.
- After the designated exposure period for oviposition, the fruit should be removed from the plant and weighed and the number and weight of fruit recorded.

2.5 Field trial design

Data to be recorded include:

- daily physical conditions (e.g. temperature, relative humidity) in the fruit holding facility
- emergence dates and numbers of all adults by species, including any abnormal adult flies.
- dates and numbers of larvae and pupae collected from the test fruit and the control fruit, noting that:
 - the medium may be sieved at the end of the holding period
 - at the end of the holding period, the fruit should be dissected before being discarded, to determine the presence of live and dead larvae or pupae; depending on the stage of fruit decay, it may be necessary to transfer the larvae to an adequate pupation medium
 - all or a subsample of pupae should be weighed and abnormalities recorded

3. Fruit Handling for Fruit Fly Development and Emergence

Fruit collected under natural conditions (surveillance by fruit sampling) and semi-natural conditions (field trials), as well as control fruit, should be kept until larval development is complete. *This period may vary with temperature and host status. Fruit handling and holding conditions should maximize fruit fly survival and be specified in the sampling protocol or experimental design of the field trial.*

Fruit should be kept in an insect-proof facility or container under conditions that ensure pupal survival, including:

- appropriate temperature and relative humidity
- suitable pupation medium.

Furthermore, conditions should facilitate accurate collection of larvae and pupae, and viable adults emerging from the fruit.

4. Data Analysis

Data from larval surveillance and field trials may be analysed quantitatively to determine, for example:

- levels of infestation (e.g. number of larvae per fruit, number of larvae per kilogram of fruit, percentage of infested fruit) at a specific confidence level
- development time of larvae and pupae, and number of viable adults
- percentage of adult emergence.

5. Record-Keeping and Publication

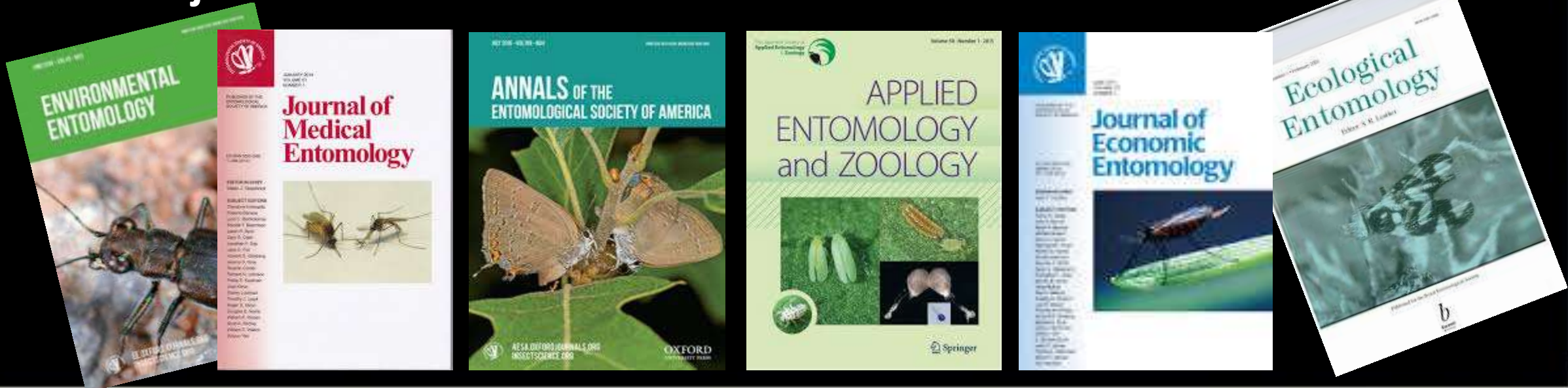
The NPPO should keep appropriate records of larval field surveillance and field trials to determine host status, including:

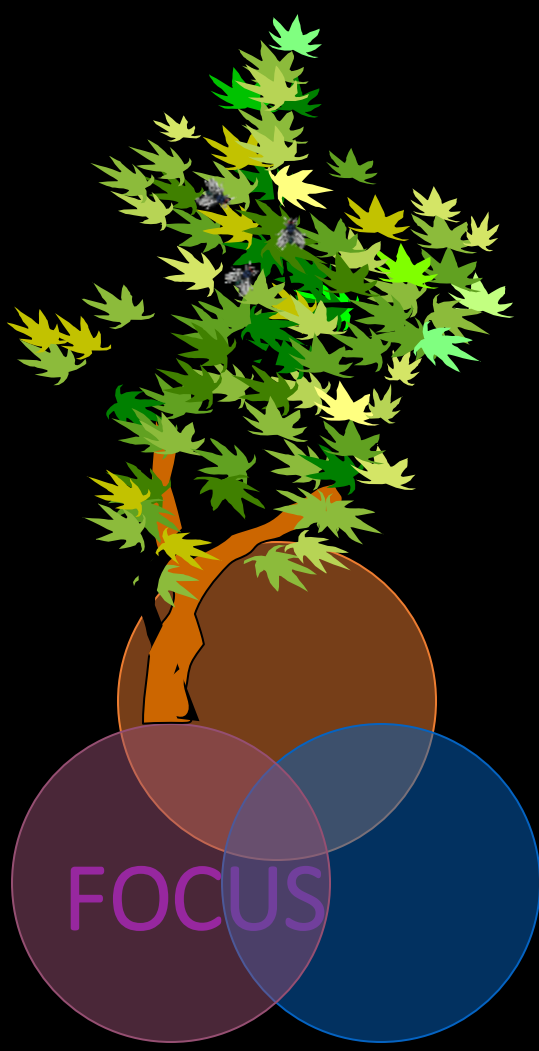
- scientific name **of the** target fruit fly
- scientific name **of the** plant species **or name of the** cultivar
- location **of the** production area **of the** fruit (including geographic coordinates)
- location **of** voucher specimens **of the** target fruit fly (to be kept in an official collection)
- origin and rearing **of the** fruit fly colony **used for the field trials**
- physical and physiological condition **of the fruit tested** for infestation **by** fruit flies
- experimental design, trials conducted, dates, locations
- raw data, statistical calculations and interpretation of results
- key scientific references used
- additional information, **including** photographs, **that may be** specific to the fruit fly, **the** fruit or host status.

5. Record-Keeping and Publication

Records **should be made** available **to the NPPO of the** importing country **upon request.**

Research **should, as far as possible, be peer reviewed and published in a scientific journal or otherwise made available.**





THANK YOU
VERY MUCH