



## Screening techniques to assess the resistance level to insect pests in rice

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### Brown Plant Hopper(BPH) and White-Backed Plant Hopper (WBPH)

#### Requirements

1. Insect rearing cages
2. Square-sized trays, Zinc trays for insect rearing and plastic trays for screening.
3. Finely powdered soil
4. Continuous maintenance of susceptible TN1 seeds, seedlings and potted plants of different age.
3. Continuous maintenance of insects throughout the screening period
4. Ideal temperature of  $30\pm 3^{\circ}\text{C}$  and humidity of 80-90% for insect maintenance.

#### Procedure

##### Preparation of screening tray

1. Make fine powder of the dry soil.
2. Fill up the tray upto  $3/4^{\text{th}}$  of the height with fine-meshed soil.
3. Make rows of uniform distance.
4. Sow test seeds in different rows along with Susceptible check TN1 and resistant check Ptb 33.
5. Cover all the rows with fine soil.
5. Sprinkle water and cover it for germination.
6. Remove the cover after germination and take care for proper growth till the plants become 10 days old.
7. Trim each row at  $10^{\text{th}}$  day of sowing by taking out seedlings so that each row will contain 25 plants.

##### Preparation for obtaining test instar(2<sup>nd</sup> instar) stage of BPH

1. Clean the BPH rearing cage to make it free from spiders, ants and other predatory organisms.
2. Place 50-60 day old potted plants of TN1 in clean Zinc trays filled to its half with water and cover each pot with a mylar or wire-mesh cage with the open end towards the top.
3. Collect gravid female BPH from the maintenance cage by sucking with an aspirator and release them in each pot through the open end of the cage @ 15 insects per pot. Release the female BPH before one day of sowing the test seeds.
4. Clean the gravid females after 48 hours of release and maintain the pots inside the cage to prevent mixing of outside BPH.
5. Observe hatching of 1<sup>st</sup> instar nymphs at 7-8 days. Allow them to remain on the potted plants for another 2 days to become 2<sup>nd</sup> instar.

### Release of test insects and observations

1. Confine the screening trays with 10 day old seedlings into insect cage which is well-cleaned.
2. Release the 2<sup>nd</sup> instar nymphs to the test tray by holding the pots upside down on the tray and gently tapping the plant so that insects will fall gently and slowly on the plants of test tray. Sucking insects by aspirator for release should be avoided as it may damage the insects.
3. Ensure that the test tray receives high insect pressure, i.e., approximately 10 insects per plant.
4. Observe everyday from the day of insect release till yellowing symptom of leaves start in susceptible variety TN1.
5. Take observation of test entries on number of plants showing yellowing symptom (damage symptom) when 95-100% plants in TN1 showed damage symptom.
6. Convert the observed data into percentage data and score them as per the standard evaluation system of IRRI, 2002. This data will represent the reaction of different test entries in mass screening. The entries with score 1 and 3, designated as highly resistant and resistant, should be further screened for confirmation.

### Confirmation of Resistance in identified donors of mass screening

1. Sow the seeds of highly resistant and resistant entries (Score 1 and 3) in replicated design with minimum of 3 replications, i.e., each entry in 3 lines along with susceptible and resistant checks.
2. Release the insect as per above procedure and allow them to feed on the plants.
3. Take mortality count of yellow/wilted/dead seedlings when all the seedlings of TN1 is affected.
4. Convert the average seedling mortality for each entry to percent data and find the score.
5. Take out healthy seedlings from score 1 entries and transplant separately for obtaining pure seed.
6. DNA Finger printing of identified donors and registration.

## Rice Gall Midge

### Requirements

1. Insect rearing cages
2. Square-sized trays for insect rearing and screening.
3. Finely powdered soil
4. Continuous maintenance of susceptible TN1 seeds and tender seedlings.
4. Continuous maintenance of insects throughout the screening period
5. Ideal temperature of  $30 \pm 3^{\circ}\text{C}$  and humidity of 80-90% for insect maintenance.

### Preparation for obtaining adult gall fly

1. Clean the rearing cage to make it free from spiders, ants and other predatory organisms.
2. Transplant 10 day-old TN1 seedlings in trays and allow them to establish for 3-4 days and then transfer the tray into insect cages.





3. Release female gall flies on the planted trays @ 15-20 / tray along with available male insects and allow the female fly to lay eggs for 2 days .
4. After 2 days, put the tray under favourable environment of  $30\pm 2^{\circ}\text{C}$  and with humidity of 80-90% and in clean and confined area so that parasites and ants will not attack the eggs or inside maggot.
5. Spray water on the rearing trays twice or more daily to ensure proper humidity which is needed for the development of the pest.
6. Observe silver shoot formation towards 18-20 days and transfer the trays to insect cages.
7. Observe the emergence of adult gall fly from silver shoot and collect them with the help of aspirator through gentle suction pressure and put them in glass test tubes @ 15 females / test tube.
8. Release insects of each test tube in a test tray prepared and kept confined in insect cages as per the method of BPH screening . After 48 hours of insect release, take care of test trays as that of maintenance trays till silver shoots are formed.
9. Record the number of silver shoot in each test entry against total plant and calculate the percent silver shoot formation in each entry.
10. Score the percent data as per SES method of IRRI(2002) and identify the highly resistant/resistant entries.
11. Repeat the screening in replicated design to confirm the reaction of highly resistant entries.
12. Transplant the highly resistant entry for pure seed collection.
13. DNA Finger printing of identified donors and registration.

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