PROBINGBEHAVIOUROFBROWNPLANTHOPPER *Nilaparvata lugens* (STAL.) IN THE RESISTANT GERMPLASM ACCESSIONS

ABSTRACT: A total of 1110 ENRRD rice germplasm accessions were evaluated for their resistance to brown planthopper,BPHinglasshousebyadoptinginternationallyacceptedstandardseedboxscreening techniqueandscoredon0-9scalebyfollowingtheIRRImethodofstandardevaluationsystemfor rice.Among1110ricegermplasmaccessionstested,3werecategorizedasHighlyresistant,24as resistant,19asmoderatelyresistant,192asmoderatelysusceptible,509assusceptibleandrestof other 363 genotypes were highly susceptible to BPH. The studies on antixenosis mechanism of resistanceforfeedingbyprobingmarksrevealedthatresistantentries,wereleastpreferredbyBPH nymphs and adults with more number of probing marks The results indicate that the germplasm accessions *viz.*, IC 343457, IC 300168 and IC 377051 were highly resistant and possess non- preference mechanism of resistance and are not preferred by BPH for feeding. The identified resistant entries can be used in the breeding programmes to develop BPH resistant varieties.

KEYWORDS:*Nilaparvatalugens*,germplasm accessions, hostplantresistance,probingmarks, feeding behavior, rice

INTRODUCTION

"Cultivation of rice isthemajoractivityandsourceofstaple food and incomeformillionsofhouseholdsroundtheglobe.InIndia,riceiscultivatedinanareaof42.96million hectares with a total production and productivityof158.75 million tonnesand 3.69metrictonnes per hectare, respectively.Paddy, isattackedbyabout800speciesofinsectpestsinbothfieldandstorage. Of all these, brown planthopper (BPH), *Nilaparvata lugens*(Stal)(Homoptera:Delphacidae)iseconomicallyimportant,whichcancausehugedamage

where both nymphs and adults suck the plant sap directly and indirectly transmits viral diseases suchasraggedstuntandgrassystunt"(Khush and Brar, 1991)."BPHdamageresultsin,appearanceofroundandyellow patches, which soon turn brownish due to the drying up of the plants which is called as 'hopper burn',andcouldresultincausingyieldlossrangingfrom10-75%.Differentchemical pesticides were advised to manage BPH, however their negligent use resulted in their revival, the development of insecticide resistance, and a negative influence on natural enemies" (Chelliah and Heinrichs, 1980., Jhansi Lakshmi *et al,* 2010a, 2010b). "An alternate strategy for controllingbrownplanthopperistoimprovehostplantresistance.Growingpest-resistantcultivars offersfarmersfreepestcontrolandcanworkwellwithotherpestcontrolstrategiesinanintegrated pest management programme. The most significant benefit of plant resistance is its particular, cumulative, and long-lasting impact on insect populations. Thenecessity to identify suitable new resistantdonorsforBPHfromdifferentsourcesisutmostimportantinordertocombatthisinsect andtodevelopmaterialresistanttodifferentbiotypes.Itisnecessarytounderstandthemechanism and factors which are responsible for manifesting the resistance into the selected cultures with desirablecharacters,sothatthesecanbeutilizedeffectivelyinthebreedingprogramme" (Jhansi Lakshmi et al, 2010a).Keeping this in view, the present investigation of identification of resistant sources from the ENRRD (EstablishmentofNationalRiceResourceDatabase)germplasmaccessionstobrownplanthopper wascarriedoutandtheantixenosismechanismofresistanceforfeedingintermsofprobingmarks was studied.

MATERIALSANDMETHODS

MassRearingofBrownPlanthopper

TheBPHwasmassrearedonsusceptiblevarietyTN1 in the Entomology greenhouse of ICAR-Indian Institute of Rice Research, Rajendranagar, Hyderabad. Twenty adult gravidfemalehopperswerereleasedonprecleanedpottedTN1plantsinovipositioncages.Plants with eggs were taken out and placed in separate cages for hatching. The hatched nymphs were utilized as and when they obtain the desired age.

MassScreening ofENRRDEntries

In order to identify the sources of resistance to BPH, 1110 ENRRD (Establishment of National Rice Resource database) germplasm accessions supplied by ICAR-National Bureau of Plant Genetic Resources, New Delhi and multiplied and maintained by ICAR-Indian institute of Rice research, Rajendranagar, Hyderabad were mass screened. Screening was carried out by adopting mass screening test under controlled greenhouse conditions as per the technique described by (Kalode *et al*, 1975, Nagendra Reddy *et al*, 2016). Theseedof the ENRRD germplasm entries waspre-germinatedinpetridishesandsown individually in the screening trays (50cm x 40cm x 8cm) filled with fertilizer enriched puddled soil. Eachscreeningtraycontained 20testlineswithabout15-20seedlingsperline,onerowofresistantcheck(PTB33)inthemiddle and two rows of susceptible check (TN1) in the boarders. Each entry was screened in two replications. After sowing, the screening trays were placed in fibre trays (60cm x 180cm x 8cm) filled with water. The screening trays werecovered withmylarcages and first and second instar BPH nymphs were released on to 12-13 day old seedlings. When more than 90 per cent plants of the susceptible check, TN1 were killed, the test entries were scored for the damage reaction, based on a 0-9 scale of International Standard Evaluation System (IRRI, 2013) as described in table 1. ENRRDgermplasmaccessionsshowingdamagescorebelow5.0weretestedwith two replications to confirm the consistency of the reaction to BPH.

Feeding/probing behaviour of third instar BPH nymphs and adults on selected ENRRD

germplasm accessions

A total number of 43 ENRRD germplasm accessions including highly resistant, resistant moderately resistant and susceptible accessions along with susceptible and resistant checks were selected to find out the feeding/probing behaviour of third instar nymphs and adults expressed in terms of feeding or probing marks on the stems of rice plants. Prior to insertion of the stylets, the planthoppersecretsasmallamountofcoagulablesalivawhilepushingthelabialtipontotheplant epidermis.Thismakesatightconnectionbetweenthemleavingcharacteristiccircularmarksatthe pount ofstylet insertion. Thesalivary deposit on theplant epidermis is called a feeding mark.The non-preference of the BPH for food was studied by counting probing marks. Number of probing marks made by a single one day old female and third instar nymph during one day feeding on resistant and susceptible rice cultures was recorded. For this purpose, a single one day old adult female insect was allowed to feed on seven day old test entry in a test tube for 24 hours and this was replicated five times. After 24 hours, the insect was removed and the test plant was stained by dipping in one per cent aqueous Erythrosin-B solution for one hour to distinguish the feeding marks on the test entries (Naito, 1964 and Ponnada, 2011). The feeding marks were counted by using magnifying hand lens. The experiment was also conducted with third instar nymphs. The data were analyzed statistically in Completely Randomized Design (CRD) and the means were separated using LSD.

Correlationandregressionanalysis:

Pearsoncorrelationanalysisandlinearregressionanalysis betweenthedamagescoreandprobingmarksofnymphsandadultswascarriedoutusingOPStat software to understand their relationship and interdependence.

RESULTSAND DISCUSSION

ENRRD germplasm accessions resistanttoBPH

Among 1110 rice germplasm accessions tested, Three entries *viz.,* IC Nos. 343457 (DS 0.3), 300168 (DS 0.6) and 377051 (DS 0.9) were highly

resistantwithadamagescoreof0.1to1.0;while,24entries*viz.*,ICNos.319799(DS1.1),343394 (DS1.2),301181(DS1.3),449821(DS1.3),343392(DS1.3),464944(DS1.4),450041(DS1.4),341334(DS 1.4),300166(DS 1.6),377527(DS 1.7),300167(DS 1.8), 319350(DS 2.1), 346927, (DS2.1),343515(DS2.2),300202(DS2.4),577624(DS2.5),545441(DS2.5),497079(DS2.7),461801(DS2.7),377423(DS2.8),554787(DS2.8),354787(DS2.9),321833(DS2.9)and252243(DS3.0),werer esistantwithadamagescoreof1.1to3.0;whereas19entries were identified as moderately resistant with a damage score of 3.1 to 5.0 (Table 2 and Fig 1). Among the remaining 1110 ENRRD entries, 192 entries were moderately susceptible (DS 5.1 to 7.0), 509 entries were susceptible (DS 7.1 to 8.9) and remaining363entrieswerehighlysusceptible(DS 9.0).PTB 33was resistant (DS 1.4)andTN1 was susceptible (DS9.0). Screeningforresistancetobrownplanthopperisacontinuousprocesstoidentify newsourcesofresistance. International Rice Research Institute (IRRI) has initiated screening of thousandsofricevarietiesandlinesincludingworldwidegermplasmcollectionsandbreedinglines ofriceinwhichalargenumberofvarieties wereidentifiedasresistant(Fernando *et al*., 1979). In India, host plant resistance to BPH is being exploited in several research centres and important sources of resistance have been identified theiruseinbreedingprogrammehasbeenattempted (Alagar *et al*, 2007,Deen *et al*, 2010, Ramulamma *et al*, 2015, Anupama *et al*, 2018 and Anjali *et al*, 2022).

Feeding/probingmarks

A total number of 41 ENRRD germplasm accessions including highly resistant and resistant accessions along with susceptible and resistant checks were selected to find out the feeding/probing behaviour of third instar nymphs and adults expressed in terms of feeding or probing marks on the stems of rice plants.

BPH adults

There was a significant difference among the entries with regard to probing marks

(Table3andFig 2).Theresistantaccessionsrecordedmorenumberofprobingmarkscomparedto susceptible accessions. Among the resistant accessions, the resistant accession, IC 377051 had maximum number of feeding punctures (20.8) whileIC145400receivedminimumnumberoffeedingpunctures(4.0). The susceptible check TN1 had 6.8 probing marks while the resistant check, PTB33 recorded the maximum number of probingmarks(44.0).The resistant germplasm accessions *viz*., IC 377051 (20.8),IC497079(19.4),IC464944(18.6),IC300167(17.8),IC319799(16.8),IC377527(16.4)recorde d more number of feeding marks compared to other resistant and susceptible entries. The germplasmaccessions*viz*.,IC145400(4.0),IC252243(4.4),IC300683(4.4),IC301172(4.4),IC 301181(4.8),IC449821 (4.8),IC 319350(4.8),IC 377746(4.8),IC343392 (5.2),IC300378(5.4)recordedverylessnumberoffeedingpunctures.Thehighlyresistantgermplasmac cessions were probed morenumber of times (14.8) by the BPH adults compared to the resistant (12.1), moderately resistant (8.1) and susceptible accessions (7.8).

BPH nymphs

There was a significant difference among the entries with regard to probing marksby nymphs (Table 2 and Fig 1). In the resistant accessions, highest number of probing marks by nymphs were observed on IC464944 (29.4) followed by IC 461801 (15.4), IC497079(12.2),IC319799(10.6)andIC450382(11.2),PTB33had the maximum number of probing marks (38.6).Lowestnumberofprobingmarkswereobserved onIC300163(2.0),followedbyIC319350(4.0),IC145400(4.2),IC343392(4.4)andIC301172(4.4)and TN1had6.8probingmarks.Nymphsprobedmorenumber of times on the highly resistant germplasm accessions (10.6) compared to resistant (10.5), moderately resistant (6.4) and susceptible entries (6.3). More number of feeding punctures were observed when the plant is fed withadultBPH(10.4)compared to the

nymphs(8.6).“Morenumberoffeedingpuncturesintheresistantentries might be due to the reason that, these resistant and moderately resistant entries did not sustain prolonged feeding due to presence of certain feeding deterrents or toxic chemicals or absence of feedingstimulants.Hence,theinsecthadtoprobemorenumberoftimesontheresistantgenotypes to locate feeding sites”(Sogawa and Pathak, 1970). The results corroborate with the findings of several workers (Ponnada *et al* 2011, Anupama *et al*., 2018, 2022, Nagendra Reddy *et al*, 2016band Priyadarsini *et al*, 2021)whoreportedthatresistantvarietiesreceivedmoreprobing marks than susceptible ones.

Correlation and regression between damage score and probing marks

Correlation analysis betweendamagescoreandprobingmarksbynymphs(-0.290^{NS})andadults(-0.303^{*})indicateda negative relation which is significant in adults. More number of probing marks were observed in the germplasm accessions which are resistant and vice versa (Table 3 and Fig 2). Anegativerelationbetweendamagescoreandprobingmarksmadebybrown planthopper and whitebackedplanthopper inricegermplasmaccessions was suggested by Ramesh *et al*, 2014 and Anupama *et al*, 2018, Priyadarsini *et al*, 2021**,** Anupama *et al*, 2022.Therewasanegativerelationbetweendamagescoreandprobingmarksand theprobingmarkscouldexplain19%variationinthedamagescoreandforeachunitincreaseintheprobing marks, the damage score is decreased by 0.328 units (Table 4 and Fig 3).

CONCLUSIONS

Inthepresentinvestigation,ENRRDgermplasm accessions*viz*.,IC343457,IC300168andIC377051werehighly resistant to brown planthopper with less damage score (0.0 to 1.0) and with more number of probingmarkswhichindicatesthattheyarenotpreferredforfeedingandtheseaccessionscanbeused in the breeding programmes to develop brown planthopper resistant varieties.

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Table1. Classificationofresistancebasedondamagereaction.

Plantstate	Damagescore	Resistanceclassification
Noneoftheleavesyellowordead	0	Highlyresistant
Onebottomleafyellow	1	Resistant
Oneortwoleavesyelloworleafdried	3	Moderatelyresistant
Oneortwoleavesdriedoroneleaf healthy	5	Moderatelyresistant
Allleavesdriedoryellowbutstem green	7	Susceptible
Plantdead	9	Highlysusceptible

Table2. Damagescoreandprobingmarksofbrownplanthopperin ENRRDgermplasmaccessions

S.No	Germplasm accessions (IC No)	DS	Probing marks made by		S.No.	Germplasm accessions (IC No)	DS	Probing marks made by	
			Nymphs	Adults				Nymphs	Adults
1	IC 343457	0.3	10.4(3.2)$^{c-g}$	8.8(2.9)$^{g-m}$	23	IC 461261	3.2	6.0(2.4)$^{e-n}$	8.4(2.9)$^{g-n}$
2	IC 300168	0.6	10.6(3.1)$^{c-g}$	14.8(3.8)$^{b-g}$	24	IC 450072	4.0	10.4(3.2)$^{c-f}$	9.2(2.9)$^{f-m}$
3	IC 377051	0.9	10.8(3.3)$^{c-e}$	20.8(4.4)^{b}	25	IC 413638	4.1	11.0(3.2)$^{c-f}$	7.4(2.7)$^{h-n}$
4	IC 319799	1.1	8.2(2.8)$^{d-m}$	16.8(3.9)$^{b-f}$	26	IC 377746	4.5	4.8(2.2)$^{j-o}$	4.8(2.1)^{mn}
5	IC 343394	1.2	6.2(2.5)$^{e-n}$	6.0(2.3)$^{j-n}$	27	IC 332672	4.6	4.8(2.2)$^{j-o}$	9.2(2.9)$^{f-m}$
6	IC 301181	1.3	6.4(2.5)$^{e-n}$	4.8(2.1)$^{l-n}$	28	IC 145400	4.6	4.2(2.0)$^{m-o}$	4.0(1.9)^{n}
7	IC 343392	1.3	4.4(2.1)$^{l-o}$	5.2(2.2)$^{k-n}$	29	IC 300378	4.7	7.6(2.7)$^{d-n}$	5.4(2.3)$^{j-n}$
8	IC 449821	1.3	6.6(2.6)$^{e-n}$	4.8(2.1) $^{l-n}$	30	IC 145402	4.7	5.2(2.2)$^{i-o}$	9.8(3.1)$^{e-k}$
9	IC 341334	1.4	10.4(3.2)$^{c-g}$	11.2(3.2)$^{d-j}$	31	IC 300683	4.8	4.6(2.1)$^{k-o}$	4.4(2.1)^{mn}
10	IC 464944	1.4	29.4(5.2)^{b}	18.6(4.1)$^{b-d}$	32	IC 300991	4.8	5.0(2.2)$^{i-o}$	10.4(3.2)$^{d-j}$
11	IC 300166	1.6	9.2(2.9)$^{d-k}$	7.6(2.7)$^{h-n}$	33	IC 450382	4.9	11.2(3.3)$^{c-e}$	9.1(2.9)$^{g-m}$
12	IC 377527	1.7	5.2(2.3)$^{i-o}$	16.4(4.0)$^{b-e}$	34	IC 17037	5.0	6.2(2.4)$^{f-n}$	13.0(3.6)$^{b-h}$
13	IC 300167	1.8	9.0(2.9)$^{d-j}$	17.8(4.2)^{bc}	35	IC 145419	5.0	8.6(2.9)$^{d-l}$	10.3(3.2)$^{d-j}$
14	IC 319350	2.1	4.0(1.9)^{no}	4.8(2.2)$^{k-n}$	36	IC 301172	5.0	4.4(2.1)$^{l-o}$	4.4(2.1)^{mn}
15	IC 343515	2.2	6.6(2.5)$^{e-n}$	11.0(3.3)$^{c-i}$	37	IC 300163	5.0	2.0(1.4)^{o}	11.0(3.2)$^{d-j}$
16	IC 300202	2.4	7.8(2.8)$^{d-n}$	7.0(2.6)$^{h-n}$	38	IC 577781	9.0	5.9(2.3)$^{g-n}$	7.4(2.6)$^{h-n}$
17	IC 545441	2.5	9.4(3.0)$^{c-i}$	9.6(3.1)$^{f-l}$	39	IC 577803	9.0	7.1(2.7)$^{d-n}$	8.0(2.8)$^{h-n}$
18	IC 577624	2.5	10.4(3.1)$^{c-h}$	12.2(3.5)$^{b-i}$	40	IC 577833	9.0	6.4(2.5)$^{e-n}$	7.0(2.6)$^{i-n}$
19	IC 461801	2.7	15.4(3.9)^{c}	11.6(3.4)$^{c-i}$	41	IC 577922	9.0	5.2(2.3)$^{h-n}$	7.4(2.7)$^{h-n}$
20	IC 497079	2.7	12.2(3.5)^{cd}	19.4(4.1)$^{b-d}$	42	PTB 33	1.4	38.6(6.1)^{a}	44.0(6.4)^{a}
21	IC 321833	2.9	5.2(2.3)$^{h-n}$	8.8(2.9)$^{g-m}$	43	TN1	9.0	6.8(2.6)$^{d-n}$	9.0(2.9)$^{g-m}$
22	IC 252243	3.0	5.8(2.3)$^{g-n}$	4.4(2.1)^{mn}					
	S.Em(±)		0.3071	0.339				0.3071	0.339
	C.V.(%)		24.84	25.07				24.84	25.07
	C.D.(0.05)		0.8573	0.9462				0.8573	0.9462

Note:Meansinacolumnfollowedbysameletterarenotsignificantlydifferentfromeachother.
Figuresin parenthesis are square root transformed values

Table3. Correlationmatrixbetweendamagescoreandprobingmarksof BPH

Componentsofresistance	Damagescore	probingmarksnymphs	probingmarksadults
Damagescore	1		
probingmarksnymphs	-0.290^{NS}	1	
probingmarksadults	-0.303*	0.404**	1

Table4. Linearregressionanalysisbetweendamagescoreandprobingmarksof BPH

Parameter	No.observations	Regressionequation	StandaradError	R^2
probingmarksNymphs	43	y=-0.1627x+12.175	0.09529726	0.1026
probingmarksAdults	43	y=-0.1652x+14.006	0.08864272	0.0915

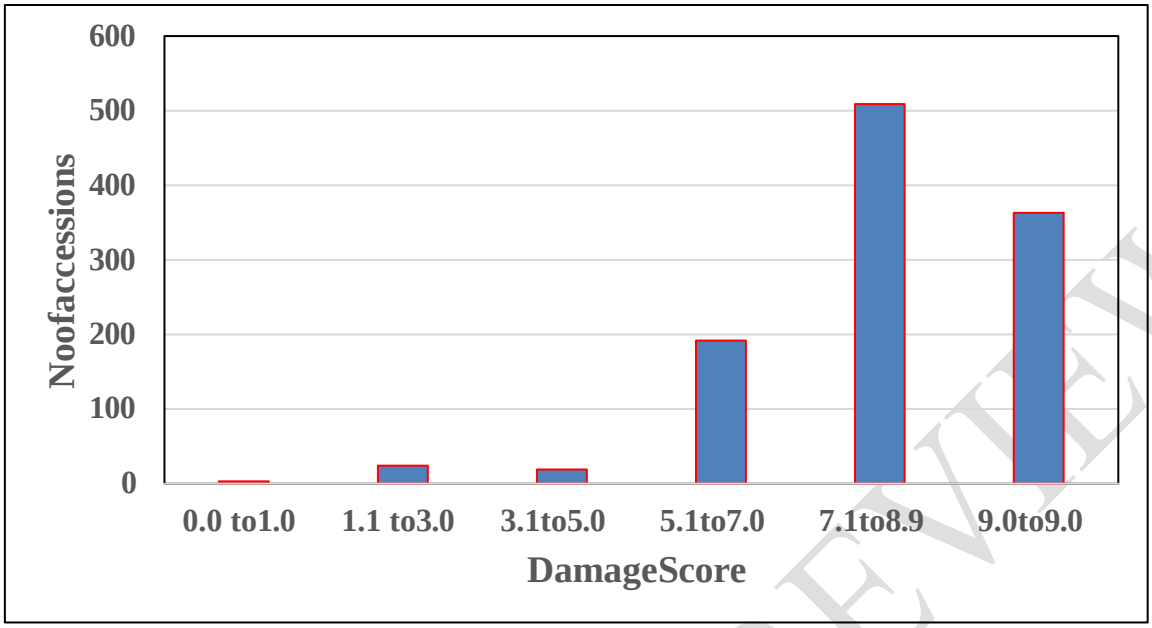

Fig1.Frequencydistribution ofdamagescorein theENRRDgermplasm accessions

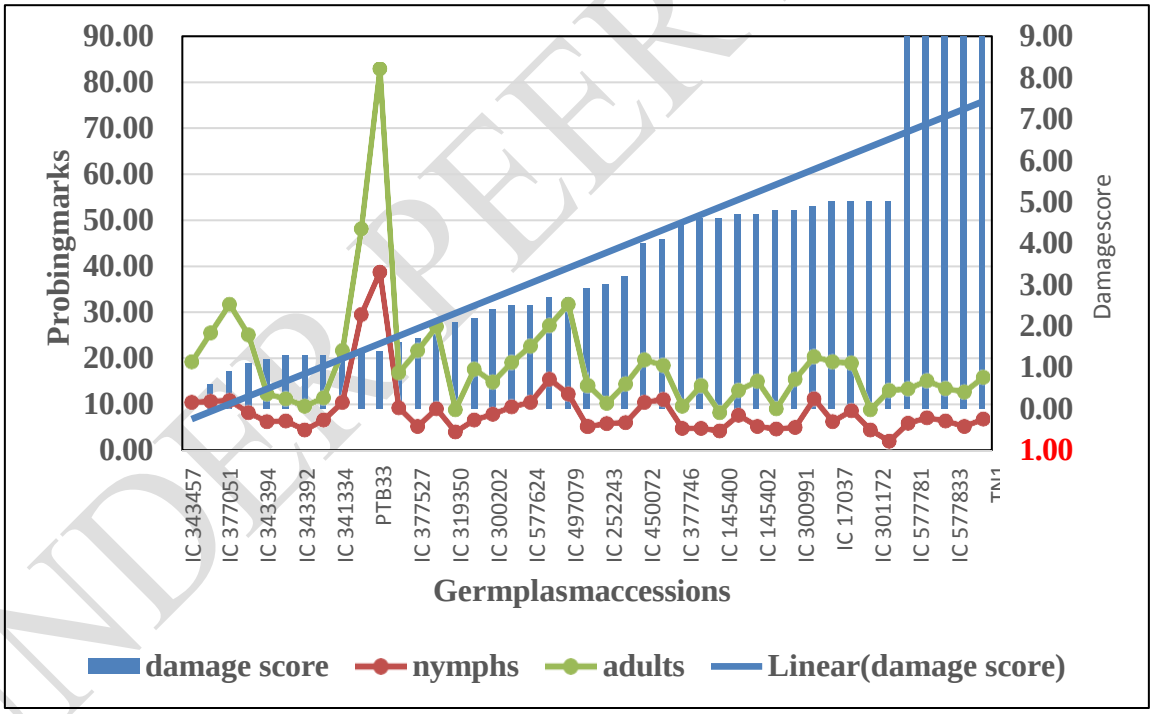

Fig2.RelationbetweendamagescoreandprobingmarksofBPH nymphsandadults

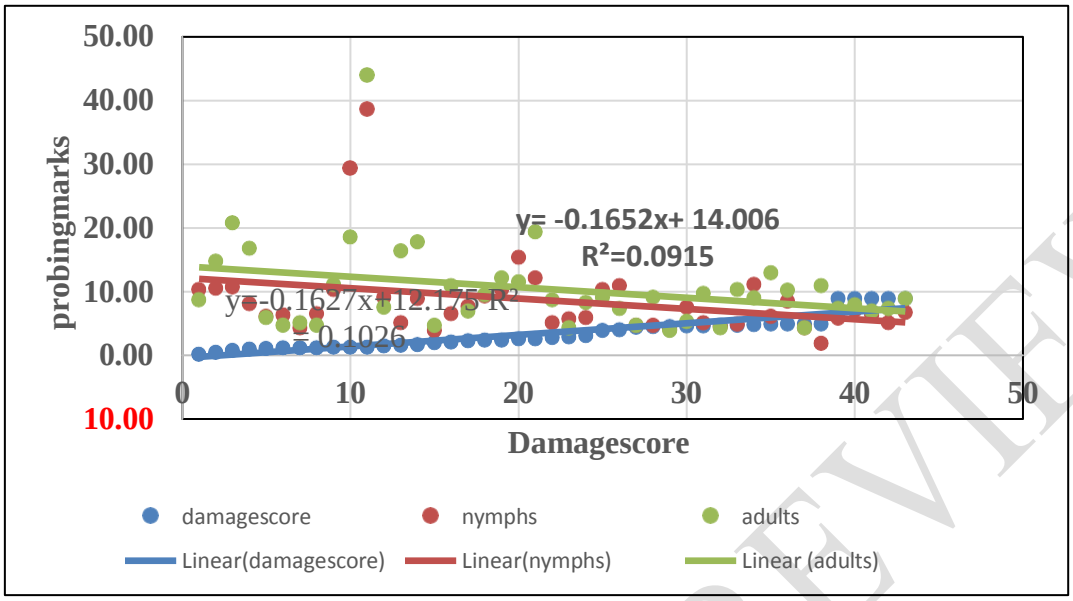

Fig3.Regressionequationbetweendamagescoreandprobingmarksof BPH

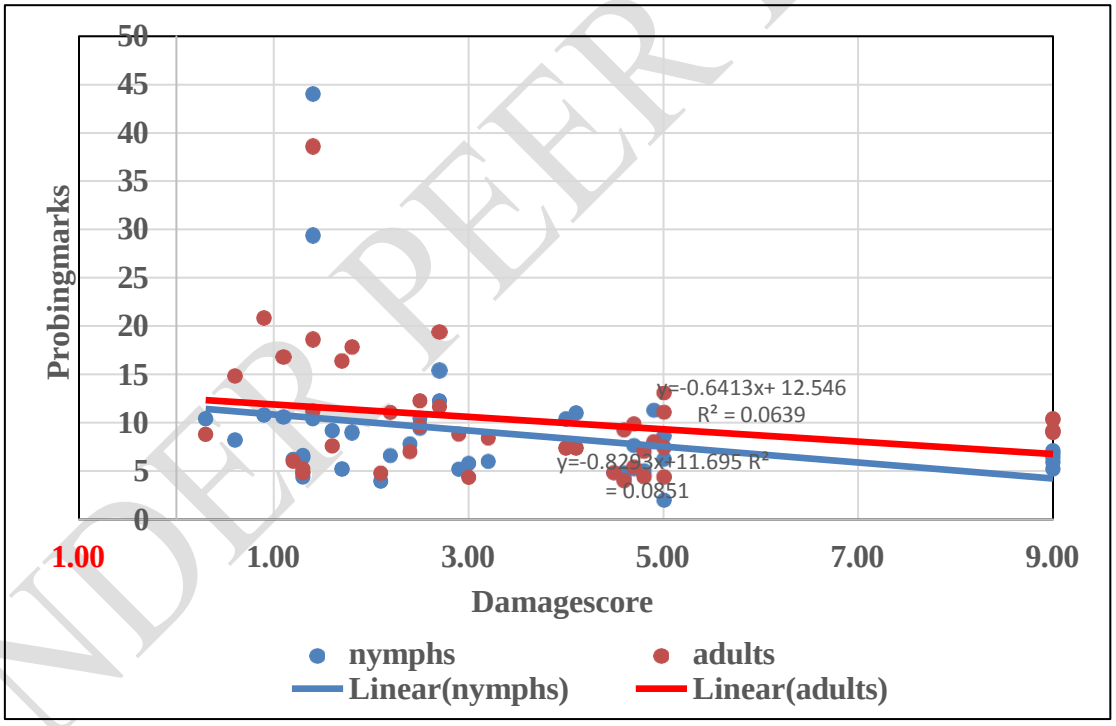

Fig4.RegressiongraphbetweendamagescoreandprobingmarksofBPH nymphsandadults

Plates1aand1b.MassscreeningofENRRDgermplasmaccessionsforBPH resistance

Plate2 Experimenton Probing marks