



Received: 23 October 2015
Accepted: 18 December 2015
Published: 08 February 2016

*Corresponding author: Firoz Hossain,
Division of Genetics, ICAR-Indian
Agricultural Research Institute (IARI),
New Delhi 110012, India
E-mail: fh_gpb@yahoo.com

Reviewing editor:
Manuel Tejada Moral, University of
Seville, Spain

Additional information is available at
the end of the article

SOIL & CROP SCIENCES | RESEARCH ARTICLE

Genetic variability among exotic and indigenous maize inbreds for resistance to stored grain weevil (*Sitophilus oryzae* L.) infestation

Rajkumar Zunjare¹, Firoz Hossain^{1*}, Vignesh Muthusamy¹, Sunil K. Jha², Pradyumn Kumar³, Javaji C. Sekhar⁴, Nepolean Thirunavukkarasu¹ and Hari S. Gupta^{5,6}

Abstract: *Sitophilus oryzae* is one of the most important stored grain pests of maize causing substantial losses. Owing to high cost, environmental and health hazards, chemical control does not provide an acceptable solution. Host plant resistance offers sustainable and cost-effective way to reduce the losses. A set of 162 diverse inbreds were evaluated to assess their responses to infestation of *S. oryzae*. Significant variation for (1) grain weight loss (GWL) (4.19–42.32%), (2) emergence of insect progeny (5.67–75.67), (3) germination of infested seeds (GIS) (0.00–81.33%), (4) pericarp thickness (PT) of the grain (47.32–194.83 μm) and (5) grain hardness (GH) (113.00–1,171.67 Newton) was observed among inbreds. GWL and emergence of insect progeny were positively correlated, while GIS was negatively correlated with both. PT and GH did not show any association with other characters. CML442 and CM394 earlier reported to be promising against *S. zeamais*, were also identified as resistant to *S. oryzae*. SKV21, Pant109, Pant124, MGB1, CML207 and LM13 were identified as resistant inbreds. These inbreds can potentially serve as valuable donors in the resistance breeding programme. The resistant inbreds can also be crossed with diverse susceptible inbreds to generate mapping populations for identifying QTLs conferring resistance to *S. oryzae*.

Subjects: Agriculture; Agriculture and Food; Crop Science; Entomology; Environment & Agriculture

Keywords: maize; stored grain; *Sitophilus oryzae*; resistance; variability

1. Introduction

Maize serves as a staple food to billions of people in Africa, Meso-America and many Asian countries (Gupta, Hossain, & Muthusamy, 2015). Together with rice and wheat, maize provides at least 30% of

ABOUT THE AUTHORS

ICAR-Indian Agricultural Research Institute, New Delhi is a premier organization for providing food- and nutritional-security in the country. The research team is involved in genetic improvement of maize for diverse biotic stresses. The present report was aimed to identify the potential sources of resistance to *Sitophilus oryzae*, a major post-harvest insect-pest that causes significant loss to maize grains in storage. The study identified promising inbreds that would serve as rich genetic resource in the resistance breeding programme.

PUBLIC INTEREST STATEMENT

The work reported in the manuscript highlights the availability of the genetic variation for weevil (*Sitophilus oryzae*) resistance in maize. The potential resistant inbreds identified in the study would serve as suitable donors in breeding strategy for imparting weevil resistance. The present work provides a sustainable and cost-effective solution to post-harvest loss of maize grains during storage.

the food calories to more than 4.5 billion people in 94 developing countries (Shiferaw, Prasanna, Hellin, & Bänziger, 2011). It contributes to >20% of total calories in human diet in 21 countries, and >30% in 12 countries. Besides, a major proportion of the maize produce is used as livestock feed, and the ever-growing poultry industry plays a major role in growth of maize especially in Asian countries. Asia alone contributes for nearly 30% of the global maize production that amounts to 1,016.73 million metric tonnes (FAOSTAT, 2013). India produced 24.35 million metric tonnes of maize during 2013–2014, and 23% of the produce is used for human food, while 63% is utilized for poultry- and animal-feed (Yadav et al., 2014).

Post-harvest losses have become one of the alarming problems worldwide, and lead to increase in food prices by removing a sizeable proportion of the produce from the supply chain that feeds to the market (Suleiman, Williams, Nissen, Bern, & Rosentrater, 2015; Zunjare, Hossain, Muthusamy, Choudhary, et al., 2015; Zunjare, Hossain, Muthusamy, Jha, et al., 2015). Among various factors, storage insect pests like grain weevils (*Sitophilus* sp.) have been recognized as the increasingly important problem across the world (Derera, Pixley, Giga, & Makanda, 2014). *Sitophilus zeamais* (maize weevil) is chiefly prevalent in Latin America, Europe and Africa, while *S. granarius* (granary weevil) is found in temperate regions. *Sitophilus oryzae*, popularly known as “rice weevil” is predominantly found in Asian countries, and is a major stored grain pest of rice and wheat, but due to polyphagous nature, it also infests maize grain particularly in Asian countries (Hossain, Boddupalli, Sharma, Kumar, & Bahadur Singh, 2007; Zunjare, Hossain, Muthusamy, Choudhary, et al., 2015; Zunjare, Hossain, Muthusamy, Jha, et al., 2015; Zunjare et al., 2014).

In tropical and subtropical regions, farmers generally store about three-fourth of the maize grains after harvest. About 12–20% grain weight loss (GWL) is common due to weevil infestation, and it may reach up to 80% under favourable conditions (Pingali & Pandey, 2001; Tefera, Demissie, Mugo, & Beyene, 2013). In developing countries like India, grains are generally stored in jute bags, unlike in the developed world, where grains are stored in commercially viable metal silos with controlled moisture and regular fumigation (Hossain et al., 2007). High humidity especially during rainy season increases the moisture content of the grain and creates conducive conditions for grain weevil infestation (Derera et al., 2014; Hossain et al., 2007). The heavy infestation leads to reduced nutritional and market-values (Masasa, Setimela, & Chiteka, 2013). Furthermore, growth of moulds on the grains due to high humidity causes mycotoxin contamination in the food and animal feed (McDonough et al., 2011).

Use of pesticides for the control of weevils does not provide a viable option for the resource poor farmers of developing countries, as it adds to the cost of cultivation. Moreover, increasing use of pesticide enhances environmental concerns viz. presence of chemical residues in food, harmful effects on non-target organisms and occurrence of resistance in insects against pesticides (Adarkwah, Obeng-Ofori, Büttner, Reichmuth, & Schöller, 2012; Nesci, Barra, & Etcheverry, 2011). Resistant genotypes on the other hand provide a practical solution to minimize losses due to insect pests, as it is cost-effective, sustainable and eco-friendly (Derera et al., 2014; Zarka, Lale, & Umeozor, 2015). Thus, studying the variability of maize genotypes in response to weevil infestation, and identification of suitable donor genotype is of prime importance for resistance breeding programme. Majority of the studies undertaken so far pertains to evaluation of maize landraces, varieties and hybrids against *S. zeamais*. Though few studies have been undertaken to evaluate maize varieties and hybrids against *S. oryzae*, no comprehensive study has been undertaken in relation to analysis of genetic variability among diverse maize inbreds. Keeping this in view, a large set of diverse inbreds of exotic and indigenous origin was characterized for their responses against *S. oryzae* with aim to: (1) study the variability of responses among inbreds to weevil infestation, (2) understand the association among different characters conferring resistance and (3) identify weevil resistant inbred(s) for their utilization in the breeding programme.

2. Materials and methods

2.1. Genetic materials

A total of 162 inbred lines comprising of 46 exotic inbreds and 116 inbreds of indigenous origin were selected to analyse their responses to infestation of *S. oryzae*. The inbreds of exotic origin comprised of genotypes from CIMMYT, Mexico (CML-), HarvestPlus Programme (HP-) and Kasetsart University, Thailand (KUI-). Indigenous inbreds were developed at various breeding centres located at different agro-ecologies of India. These inbreds were evaluated for their responses against *S. oryzae* in two experiments viz. Experiment-I (50 inbreds) and Experiment-II (112 inbreds). In absence of suitable resistant checks against *S. oryzae*, CML394 and CML442 found promising against *S. zeamais* were used as putative resistant checks in Experiments I and II, respectively (Dhliwayo & Pixley, 2003). MGQSO370 and CM140 were used as susceptible control in the experiments I and II, respectively (Hossain et al., 2007). The experiments were undertaken in a completely randomized design with three replications, each having 25 kernels per genotype. Characters namely (1) grain weight loss (GWL), (2) number of insect progeny emerged (NIP), (3) germination of infested seeds (GIS), (4) pericarp thickness (PT) and (5) grain hardness (GH) were recorded.

2.2. Screening technique for weevil infestation

Twenty-five kernels from each replication were wrapped in a cotton cloth and put in desiccators with 25% KOH solution, for 20 days to condition the seeds at ~15% moisture. The conditioned kernels were weighed, and transferred to a plastic petriplate with ventilated upper lid. Eight pairs of newly emerged unsexed adult insects were kept for seven days in each of the petri plates (García-Lara, Khairallah, Vargas, & Bergvinson, 2009; Hossain et al., 2007). Insects were allowed to mate; the eggs are laid just beneath the pericarp of the kernel. After seven days, the released insect pairs were taken out, and the petri plates with 25 kernels were kept in a biological oxygen demand (BOD) incubator at $28 \pm 2^\circ\text{C}$ and $70 \pm 5\%$ Relative Humidity (RH). After 30 days in the BOD incubator, the petri plates were monitored regularly for the emergence of insect progenies. After the emergence of first weevil, progenies were counted and removed from each petri plate on every alternate day for a period of next 40 days (Hossain et al., 2007). After the completion of the experiment, GWL was calculated using $[(\text{Initial weight} - \text{final weight})/\text{initial weight}] \times 100$. NIP per genotype was computed by adding the number weevils emerged during 40-days period (Zunjare, Hossain, Muthusamy, Choudhary, et al., 2015; Zunjare, Hossain, Muthusamy, Jha, et al., 2015; Zunjare et al., 2014).

2.3. Germination of infested seeds

After the completion of infestation experiments, each of the petri plates was filled with fertile soil having fine tilth. Adequate water was applied as and when required and kept for next 10 days at $25\text{--}30^\circ\text{C}$. Percentage germination was then calculated as number of plants germinated over 25 kernels kept for germination.

2.4. Pericarp thickness

Three randomly selected kernels per replication were used to measure PT using compound microscope with ocular and stage micrometres. 2.5 g of safranin stain in 100 ml of 95% ethanol was used to make a stock solution. Kernels were soaked for 24 h, and crown of the kernel was removed using a single-edged razor. Free hand section on the abgerminal side of the kernel was selected and was stained with dilute safranin. PT of the section was measured under power (10 \times) using an ocular micrometre (Tracy, Chandravadana, & Galinat, 1978). It was found that 1 ocular unit was equal to 0.0167 mm on the stage micrometre scale. PT (μm) was measured as ocular units \times 16.7.

2.5. Grain hardness

GH was determined by Texture Analyzer (Model: TA + Di, Scientific Microsystem, UK). The hardness was measured in non-infested grains after conditioning at ~15% moisture. For measurement of hardness, a cylindrical probe of 75-mm diameter was used. The test speed of the probe was fixed at 2 mm/s and the compression distance at 70%. The load cell of 500-kg capacity was used. The first

peak force (N: Newton) in the force deformation curve was noted as GH. The GH of five grains per genotype was used to calculate average hardness (Mohsenin, 1980).

2.6. Statistical analyses

The GWL was angular transformed as per the procedure given by Bliss (1937). Pearson’s simple correlations among characters were computed using MS Office-Excel 2007. The data were analysed for analyses of variance (ANOVA) using SAS Version 6.12. A cumulative resistance index (CRI) for each of the genotypes was computed using least significant difference (LSD) rank, with equal weight for each of characters contributing to resistance (Arunachalam & Bandopadhyay, 1984). The groups of genotypes calculated as per the LSD was used to compute a value for a specific genotypes, and values across characters were then added to create CRI score.

3. Results

3.1. Genetic variability

ANOVA revealed significant variation in GWL, NIP, GIS, PT and GH across experiments (Table 1). GWL ranged from 4.50 to 42.32% (mean: 19.48%) and 4.19 to 42.19% (mean: 16.28%) among the genotypes evaluated in Experiment-I and -II, respectively. CML394 (4.50%), SKV21 (5.43%), V400 (6.53%), LM13 (7.00%) and V351 (7.47%) were found promising with a low GWL in Experiment-I. In Experiment-II, CML207 (4.19%), Pant124 (4.4%), CM502 (5.52%), Pant109 (6.17%) and CML442 (6.79%) showed low GWL. NIP in Experiment-I ranged from 5.67 to 69.33 (mean: 26.02), while the same varied from 6.00 to 75.67 (mean: 24.21) in Experiment II. LM11 (5.67), LM13 (7.00), SKV21 (7.00), CML394 (7.33) and Pant112 (7.67) were identified as promising with low NIP in Experiment-I. In Experiment-II, Pant124 (6.00), Pant109 (6.00), V334 (6.67), CML442 (7.33), BAJIM-06-17 (7.67) and HUZM185 (7.67) recorded low NIP. GIS exhibited wide variation in both Experiment-I (range: 0.00–81.33%; mean: 23.49%) and Experiment-II (range: 0.00–77.33%; mean: 30.17%). CML394 (81.33%), SKV21 (76.00%), V351 (70.67%) and LM13 (70.67%) in Experiment-I showed higher GIS. In Experiment-II, highest GIS (77.33%) was recorded in CM502, followed by 73.33% (MGB1 and CML61), 72.00% (BAJIM-06-17, HKI209, BAJIM-06-15 and MGHC2), 70.67% (Pant109, V334, CML200 and MGHC1) and 69.33% (CML442, V372 and CML288).

Average PT among the inbreds was 104.41 µm and 102.18 µm with a range from 58.45 to 189.27 µm and 47.32 to 194.83 µm in Experiment-I and -II, respectively. Of the 50 inbreds studied in Experiment-I, HKI1344 (189.27 µm) recorded highest PT, followed by V351 (172.57 µm), CML436 (153.08 µm) and SKV21 (150.30 µm). Resistant check CML394 possessed PT of 94.63 µm, while the susceptible check, MGQSO370 had a PT of 97.42 µm. In Experiment-II, inbreds with higher PT include CML465 (194.83 µm), V364 (172.57 µm), BAJIM-06-17 (164.22 µm) and CML200 (161.43 µm). The susceptible check CM140 showed PT of 155.87 µm, and resistant check, CML442 possessed 97.42 µm of PT.

Table 1. ANOVA depicting mean sum of square for different characters studied for weevil infestation in Experiment-I and -II

Source of variation	df		MSS									
			GWL		NIP		GIS		PT		GH	
	Exp-I	Exp-II	Exp-I	Exp-II	Exp-I	Exp-II	Exp-I	Exp-II	Exp-I	Exp-II	Exp-I	Exp-II
Genotypes	49	111	197.35**	176.72**	645.84**	663.29**	1,859.89**	2,116.34**	2,450.15**	2,314.43**	63,202.4**	60,934.4**
Error	100	224	9.25	7.88	21.40	18.77	94.51	110.24	120.18	91.66	3,325.10	4,505.2
Total	149	335	-	-	-	-	-	-	-	-	-	-

**Significant at $p = 0.01$.

Notes: df: degrees of freedom; MSS: Mean sum of square; GWL: grain weight loss; NIP: number of insect progeny emerged; GIS: germination of infested seeds; PT: pericarp thickness; GH: grain hardness; Exp-I: Experiment-I; Exp-II: Experiment-II.

GH among inbreds of Experiment-I, varied from 113.0 to 834.67 N with a mean of 301.57 N. In Experiment-II, 136.00–1,171.67 N was observed with a mean GH of 335.59 N. MGHC5 (834.67 N), CML466 (720.67 N) and CML436 (703.33 N) of Experiment-I, and CI4 (1,171.67 N) and BAJIM-10-01 (783.00 N) of Experiment-II recorded high GH. Many inbreds recorded much higher GH compared to resistant checks CML394 (452.67 N) and CML442 (235.67 N). Susceptible checks, CM140 and MGQSO370 recorded GH of 368.00 N and 113.00 N, respectively.

3.2. Association among characters

Correlation analyses revealed strong positive association between GWL and NIP in Experiment-I ($r = 0.88$) and -II ($r = 0.82$) (Table 2). GWL was negatively correlated with GIS in Experiment-I ($r = -0.74$) and -II ($r = -0.83$). Similar association ($r = -0.66$ and -0.74) between GIS and NIP was also observed in Experiment-I and -II, respectively. PT showed weak negative association with GWL ($r = -0.28$) in Experiment-I, while it did not exhibit any correlation in Experiment-II. GH did not possess any association with either of GWL and NIP across experiments (Table 2). Based on the above results, GWL, NIP and GIS were considered, while PT and GH were not considered for calculating the CRI.

3.3. Cumulative resistance index

CRI exhibited wide variation among inbreds evaluated in both the experiments. In Experiment-I, CRI ranged from 0.10 to 2.93 with a mean of 1.36. Experiment-II showed variation for CRI from 0.05 to 2.89, with a mean of 1.52. Higher value of CRI depicts resistance, while low value indicates its susceptibility to *S. oryzae* (Zunjare et al., 2014). Resistant checks, CML394 and CML442 had very high CRI, while susceptible checks, MGQSO370 and CM140 possessed very low CRI (Table 3). Among the inbreds, SKV21, LM13, Pant109, Pant124, MGB1 and CML207 possessed high CRI.

4. Discussion

The present study demonstrated the existence of wide genetic variation for GWL (4.19–42.32%) and NIP (5.67–75.67) across experiments. Hossain et al. (2007) evaluated maize genotypes against *S. oryzae* and reported GWL ranging from 11.00 to 41.00%. A range of 10.25–97.75 for NIP was reported by De and Sarup (1990) in a set of maize composites and hybrids against *S. oryzae*. Hossain et al. (2007) found the similar type of variation (5.70–59.30) for NIP. Significant genetic variation for these two characters has also been recorded against *S. zeamais* (Dari, Pixley, & Setimela, 2010; Derera et al., 2014; García-Lara et al., 2009; Masasa et al., 2013). The present study showed that GWL and NIP were also positively correlated with each other. This is due to the fact that female weevil prefer seeds of susceptible genotypes compared to resistant inbreds for egg laying (Kim & Kossou, 2003). Larvae after hatching start eating the inside of the seed for their development, and the extent

Table 2. Correlation coefficients among characters studied for weevil infestation in Experiment-I and -II

Experiments	Characters	NIP	GIS	PT	GH
Exp-I	GWL	0.88**	-0.74**	-0.28*	-0.04
Exp-II		0.82**	-0.83**	-0.03	0.02
Exp-I	NIP		-0.66**	-0.18	-0.06
Exp-II			-0.74**	0.01	0.04
Exp-I	GIS			0.02	0.09
Exp-II				-0.01	-0.07
Exp-I	PT				0.16
Exp-II					0.08

*Significant at $p = 0.05$.

**Significant at $p = 0.01$.

Notes: GWL: grain weight loss; NIP: number of insect progeny emerged; GIS: germination of infested seeds; PT: pericarp thickness; GH: grain hardness; Exp-I: Experiment-I; Exp-II: Experiment-II.

Table 3. Responses of promising inbreds and their comparison with checks against infestation of *S. oryzae*

S. No.	Genotype	Source	GWL (%)	NIP	GIS (%)	PT (µm)	GH (Newton)	CRI
1.	CML394 (Resistant check)	CIMMYT, Mexico	4.50	7.33	81.33	94.63	452.67	2.93
2.	SKV21	UAS-Nagena-halli	5.43	7.00	76.00	150.30	356.33	2.89
3.	Pant109	GBPUAT, Pantnagar	6.17	6.00	70.67	71.25	321.00	2.89
4.	Pant124	GBPUAT, Pantnagar	4.40	6.00	64.00	109.11	371.00	2.88
5.	CML442 (Resistant check)	CIMMYT, Mexico	6.79	7.33	69.33	97.42	235.67	2.85
6.	CML207	CIMMYT, Mexico	4.19	8.00	64.00	83.50	404.67	2.83
7.	MGB1	IARI, New Delhi	7.29	8.33	73.33	105.77	372.67	2.83
8.	HUZM185	BHU, Varanasi	7.48	7.67	66.67	102.98	264.67	2.79
9.	HKI209	CCSHAU, Uchani	7.89	8.00	72.00	83.50	233.67	2.79
10.	LM13	PAU, Ludhiana	7.00	7.00	70.67	122.47	386.67	2.78
11.	MGHC1	IARI, New Delhi	7.28	10.00	70.67	125.25	323.67	2.75
12.	BAJIM-06-15	CSK-HPKV, Bajaura	7.94	9.00	72.00	114.12	216.00	2.75
13.	MGQSO370 (Susceptible check)	IARI, New Delhi	36.54	50.33	0.00	97.42	113.00	0.16
14.	CM140 (Susceptible check)	PAU, Ludhiana	25.07	35.00	2.67	155.87	368.00	0.45

Notes: CRI: Cumulative Resistance Index; GWL: grain weight loss; NIP: number of insect progeny emerged; GIS: germination of infested seeds; PT: pericarp thickness; GH: grain hardness.

of seed damaged is more in susceptible inbreds, while it is less in resistant ones (Dari et al., 2010; Derera et al., 2014; Garcia-Lara & Bergvinson, 2014; Masasa et al., 2013). Endosperm of susceptible inbred promotes growth of the larvae much faster and in turn results into more GWL as compared to resistant genotypes (Castro-Álvarez, William, Bergvinson, & García-Lara, 2015). The GWL and NIP are positively correlated to *S. zeamais* infestation, and have been included in calculating susceptibility index worldwide (Dari et al., 2010; García-Lara et al., 2009; Siwale, Mbata, Mrobert, & Lungu, 2009). Hossain et al. (2007) and Zunjare et al. (2014) also reported the role of GWL and NIP in imparting resistance in maize inbreds evaluated against *S. oryzae*.

The viability of maize seed is reported to be drastically affected by weevil infestation (Caneppele, Caneppele, Lázari, & Lázari, 2003; Okiwelu, Adu, & Okonkwo, 1987). Weevils while causing internal seed damage affect seeds' physiological quality and embryo leading to direct reduction in germination (Caneppele et al., 2003). In the present study, weevil infestation drastically affected the GIS, with no germination in many of the genotypes. Okiwelu et al. (1987) reported GIS being varied from 43.00 to 97.50%, while evaluating the genotypes with different dose of insect infestation. Santos, Maria, and Cruz (1990) conducted germination test to determine effect of maize weevil, at various stages of insect development and showed that germination reduced proportionally with insect growth. De and Sarup (1990) also reported a variation of 4.00–72.00% for germination after weevil (*S. oryzae*) infestation in selected maize composites and hybrids. Among the various specialty corn inbreds, QPM and sweet corn lines showed drastic loss of GIS than popcorn lines (Zunjare et al.,

2014). The present study revealed strong negative correlation between GIS with both GWL and NIP. A susceptible genotype possesses more NIP, suggesting that more number of eggs per seed are laid on susceptible inbred. Further, more GWL indicates that the developing larvae consume more quantity of seed endosperm. The damage caused to the embryo directly reduces the germination, while the loss of endosperm fails to provide the requisite vigour to the growing embryo during germination, thereby indirectly reducing the germination percentage (Yan, Duermeyer, Leoveanu, & Nambara, 2014). Further, adult weevils sometime enter the seed through soft embryo for egg laying and cause irreversible damage (Caneppele et al., 2003). The inbreds having damaged embryo show substantial reduction in germination. In the present study, despite weevil infestation some of the inbreds recorded >70% germination, thereby suggesting their resistance nature.

The inbreds revealed wide variation for PT (47.32–194.83 μm). However, PT did not reveal strong association with GWL and NIP. PT was comparable among resistant check, CML394 (94.63 μm) and susceptible check, MGQSO-370 (97.42 μm) in Experiment-I and in Experiment-II, the resistant check, CML442 (97.42 μm) had much less PT than the susceptible check, CM140 (155.87 μm). Thus, PT may not be a key factor for imparting resistance against weevils. This finding was also supported by the results of Gomez, Rodriguez, Poneleit, and Blake (1983), where they did not find any significant association of PT with weevil resistance. Recently, Zunjare et al. (2014) demonstrated non-significance of PT in conferring weevil resistance. In contrast, significant positive correlation between PT and weevil resistance has been reported (Arnason et al., 1992; Bergvinson, 2001). Snout penetration by the weevils into the grains depends on the thickness of the pericarp, and grains with thick pericarp are harder in nature and are very tough to penetrate (Kelvin, 2002).

Kelvin (2002) reported that the snout penetration by the weevils into the grain depends on the hardness of the kernel. The present study revealed significant variation for GH (113.0–1,171.67 N), though it did not show any association with either of GWL and NIP. García-Lara et al. (2009) and Dari et al. (2010) while working with *S. zeamais* demonstrated weak association between the GH and weevil resistance. Properly dried popcorn grains are much harder as compared to other type of inbreds, while QPM grains are relatively soft due to the presence of *opaque2* allele (Vasal, Villegas, Bjarnason, Gelaw, & Goertz, 1980). Zunjare et al. (2014) analysed a set of specialty corn inbreds including popcorn, sweet corn (shrunken and sugary types) and QPM inbreds against *S. oryzae* infestation, and found no association between GH with level of infestation. On the contrary, GH has been identified as an important mechanism of resistance against the maize weevil (Arnason et al., 1992; Bergvinson, 2001; García-Lara & Bergvinson, 2014). Siwale et al. (2009) reported that GH is closely related to maize weevil resistance. Ruswandi, Nono, Susanto, and Puspita (2009) and Bergvinson (2001) also reported negative correlation between GH and grain damage. Since physical barriers such as PT and GH did not contribute to weevil resistance in maize in the present study, biochemical properties such as α -amylase and protease-inhibitors, and phenolics such as E-ferulic acid may explain the basis of resistance to weevil infestation in maize (Arnason et al., 1992; Classen et al., 1990; García-Lara & Bergvinson, 2014). CML394 and CML442 were earlier reported as resistant inbreds against *S. zeamais* by Dhliwayo and Pixley (2003). We also found both of them to be promising against *S. oryzae*, thereby indicating that mechanism of resistance against *S. zeamais* and *S. oryzae* may not differ much.

GWL and NIP have been traditionally used to formulate index for conferring resistance or susceptibility (Derera et al., 2014; Dhliwayo & Pixley, 2003; García-Lara et al., 2009). GIS has not been taken into consideration for computing the index, as harvest (F_2 grains) of the F_1 hybrids are not used for sowing in the next season, instead consumed as food and animal feed. However, parental inbreds of the hybrids are required to be stored properly every year for generation of F_1 seeds in the next season. Infestations by weevil should not only cause less damage to the parental inbred seeds, but also have acceptable level of germination. Further, inbreds are also used to develop synthetics/composite, and seeds are stored for generation of same composite in the next year. Thus, seeds of the composites should have proper germination even after infestation by the weevils. Zunjare et al. (2014) observed that average GWL and NIP in shrunken (GWL: 12.64%; NIP: 9.66) and sugary (GWL:

12.09%, NIP: 12.47) inbreds were comparable to popcorn inbreds (GWL: 8.19%, NIP: 9.23). However, popcorn inbreds recorded average 78.28% GIS compared to 21.41 and 21.47% in shrunken and sugary inbreds, respectively. This demonstrated that computing index only based on GWL and NIP may be misleading, and GIS also plays vital role. In the present study, GIS was thus taken into consideration with GWL and NIP for computing the CRI. Across experiments, CML394, CML442, SKV21, Pant109, Pant124, MGB1, CML207 and LM13 were found to be resistant with high CRI value. These inbreds serve as valuable genetic resource in the resistance breeding programme. These inbreds can be crossed to generate hybrid combinations possessing resistance against *S. oryzae* in maize. Besides, gene pool can be created by ensuring random mating among the resistant inbreds, and the population thus generated can be used for deriving new inbred lines with resistance to weevil infestation. Genetic analyses against *S. zeamais* revealed that additive gene action is an important fraction of the genetic variance (Dhliwayo & Pixley, 2003; Kanyamasoro, Karungi, Asea, & Gibson, 2012). Thus, crossing diverse resistant inbreds would likely to produce transgressive segregants in F_2 and inbreds with higher degree of resistance can be developed (Castro-Álvarez et al., 2015). Further, genetic loci contributing resistance to *S. oryzae* have not been identified so far. Thus, resistant inbreds can be crossed with highly susceptible inbreds viz. V401 (CRI = 0.05), LM17 (CRI = 0.08), CM144 (CRI = 0.08), BAJIM-08-10 (CRI = 0.10) to generate mapping populations for identifying QTLs conferring resistance to *S. oryzae* infestation.

4. Conclusions

The present study dealt with the comprehensive evaluation of diverse maize inbreds against infestation of *S. oryzae*. Wide natural variation for GWL, NIP, GIS, PT and GH exists among the inbreds. Correlation analyses revealed that GWL, NIP and GIS are the contributory characters for imparting resistance, while PT and GH did not impart resistance against weevil infestation. Based on CRI, promising inbreds with higher degree of resistance to *S. oryzae* have been identified, and can be potentially utilized as donors in resistance breeding programme. Mapping populations generated among the contrasting inbreds would help in identifying QTLs conferring resistance to weevil infestation.

Acknowledgements

The financial support from the Indian Council of Agricultural Research in the form of Junior Research Fellowship awarded for first author's M.Sc. programme is gratefully acknowledged. We also thank the breeders of CIMMYT, HarvestPlus, Kasetart University and maize breeding centres of All India Coordinated Maize Improvement Project, India for sharing their inbred lines.

Funding

The authors received no direct funding for this research.

Competing interests

The authors declare no competing interest.

Author details

Rajkumar Zunjare¹
E-mail: raj_gpb@yahoo.com
ORCID ID: <http://orcid.org/0000-0001-5001-2681>
Firoz Hossain¹
E-mail: fh_gpb@yahoo.com
Vignesh Muthusamy¹
E-mail: pmvignesh@yahoo.co.in
ORCID ID: <http://orcid.org/0000-0003-3169-890X>
Sunil K. Jha²
E-mail: skj_ageg@iari.res.in
Pradyumn Kumar³
E-mail: pradyumn.kumar@gmail.com
ORCID ID: <http://orcid.org/0000-0002-0202-0350>
Javaji C. Sekhar⁴
E-mail: javajics@gmail.com
Nepolean Thirunavukkarasu¹
E-mail: tnepolean@gmail.com

ORCID ID: <http://orcid.org/0000-0002-6263-539X>

Hari S. Gupta^{5,6}

E-mail: hsgupta.53@gmail.com

¹ Division of Genetics, ICAR-Indian Agricultural Research Institute (IARI), New Delhi 110012, India.

² Division of Food Science and Post Harvest Technology, ICAR-Indian Agricultural Research Institute (IARI), New Delhi 110012, India.

³ ICAR-Indian Institute of Maize Research (IIMR), New Delhi 110012, India.

⁴ Winter Nursery Centre, ICAR-Indian Institute of Maize Research (IIMR), Hyderabad 500030, India.

⁵ ICAR-Indian Agricultural Research Institute (IARI), New Delhi 110012, India.

⁶ Borlaug Institute for South Asia (BISA), New Delhi 110012, India.

Citation information

Cite this article as: Genetic variability among exotic and indigenous maize inbreds for resistance to stored grain weevil (*Sitophilus oryzae* L.) infestation, Rajkumar Zunjare, Firoz Hossain, Vignesh Muthusamy, Sunil K. Jha, Pradyumn Kumar, Javaji C. Sekhar, Nepolean Thirunavukkarasu & Hari S. Gupta, *Cogent Food & Agriculture* (2016), 2: 1137156.

References

Adarkwah, C., Obeng-Ofori, D., Büttner, C., Reichmuth, C., & Schöller, M. (2012). Potential of *Lariophagus distinguendus* (Förster) (Hymenoptera: Pteromalidae) to suppress the maize weevil *Sitophilus zeamais* Motschulsky (Coleoptera: Curculionidae) in bagged and bulk stored maize. *Biological Control*, 60, 175–181.
<http://dx.doi.org/10.1016/j.biocontrol.2011.11.003>

- Arnason, J., Gale, J., Conilh de Beysac, B., Sen, A., Miller, S., Philogene, B., ... & Mihm, J. (1992). Role of phenolics in resistance of maize grain to the stored grain insects, *Prostephanus truncatus* (Horn) and *Sitophilus zeamais* (Motsch.). *Journal of Stored Products Research*, 28, 119–126.
[http://dx.doi.org/10.1016/0022-474X\(92\)90019-M](http://dx.doi.org/10.1016/0022-474X(92)90019-M)
- Arunachalam, V., & Bandopadhyay, A. (1984). A method to take decisions jointly on a number of different characters. *Indian Journal of Genetics and Plant Breeding*, 43, 419–424.
- Bergvinson, D. J. (2001). Storage pest resistance in maize. In: *Maize program. Maize Research highlights 1999–2000* (pp. 32–39). Mexico D.F.: CIMMYT.
- Bliss, C. I. (1937). Analysis of field experimental data expressed in percentage. *Plant Protection*, 12, 67–77.
- Caneppele, M. A. B., Caneppele, C., Lazzari, F. A., & Lazzari, S. M. N. (2003). Correlation between the infestation level of *Sitophilus zeamais* Motschulsky, 1855 (Coleoptera, Curculionidae) and the quality factors of stored corn. *Zea mays* L. (Poaceae). *Revista Brasileira de Entomologia*, 47, 625–630. <http://dx.doi.org/10.1590/S0085-56262003000400015>
- Castro-Álvarez, F. F., William, M., Bergvinson, D. J., & García-Lara, S. (2015). Genetic mapping of QTL for maize weevil resistance in a RIL population of tropical maize. *Theoretical Applied Genetics*, 128, 411–419.
<http://dx.doi.org/10.1007/s00122-014-2440-6>
- Classen, D., Arnason, J. T., Serratos, J. A., Lambert, J. D. H., Nozzolillo, C., & Philogene, B. J. R. (1990). Correlation of phenolic acid content of maize to resistance to *Sitophilus zeamais*, the maize weevil, in CIMMYT's collections. *Journal of Chemical Ecology*, 16, 301–315.
<http://dx.doi.org/10.1007/BF01021766>
- Dari, S., Pixley, K. V., & Setimela, P. (2010). Resistance of early generation maize inbred lines and their hybrids to maize weevil [(Motschulsky)]. *Crop Science*, 50, 1310–1317.
<http://dx.doi.org/10.2135/cropsci2009.10.0621>
- De, D., & Sarup, P. (1990). Identification of resistant maize varieties to save losses in storage due to *Sitophilus oryzae* (L.). *Journal of Entomological Research*, 14, 63–73.
- Derera, J., Pixley, K. V., Giga, D. P., & Makanda, I. (2014). Resistance of maize to the maize weevil: III. Grain weight loss assessment and implications for breeding. *Journal of Stored Products Research*, 59, 24–35.
<http://dx.doi.org/10.1016/j.jspr.2014.04.004>
- Dhliwayo, T., & Pixley, K. V. (2003). Divergent selection for resistance to maize weevil in six maize populations. *Crop Science*, 43, 2043–2049.
<http://dx.doi.org/10.2135/cropsci2003.2043>
- FAOSTAT. (2013). *Food and Agriculture Organization of the United Nations Statistical Database results*. Retrieved from <http://www.fao.org/>
- García-Lara, S., & Bergvinson, D. J. (2014). Phytochemical changes during recurrent selection for storage pest resistance in tropical maize. *Crop Science*, 54, 1–10.
- García-Lara, S., Bergvinson, D. J., Burt, A. J., Ramputh, A. I., Díaz-Pontones, D. M., & Arnason, J. T. (2004). The role of pericarp cell wall components in maize weevil resistance. *Crop Science*, 44, 1560–1567.
- García-Lara, S., Khairallah, M. M., Vargas, M., & Bergvinson, D. J. (2009). Mapping of QTL associated with maize weevil resistance in tropical maize. *Crop Science*, 49, 139–149.
<http://dx.doi.org/10.2135/cropsci2007.06.0326>
- Gomez, L. A., Rodríguez, J. G., Poneleit, C. G., & Blake, D. F. (1983). Relationship between some characteristics of the corn kernel pericarp and resistance to the rice weevil (Coleoptera: Curculionidae). *Journal of Economic Entomology*, 76, 797–800.
<http://dx.doi.org/10.1093/jee/76.4.797>
- Gupta, H. S., Hossain, F., & Muthusamy, V. (2015). Biofortification of maize: An Indian perspective. *Indian Journal of Genetics and Plant Breeding (The)*, 75, 1–22.
<http://dx.doi.org/10.5958/0975-6906.2015.00001.2>
- Hossain, F., Boddupalli, P. M., Sharma, R. K., Kumar, P., & Bahadur Singh, B. (2007). Evaluation of quality protein maize genotypes for resistance to stored grain weevil *Sitophilus oryzae* (Coleoptera: Curculionidae). *International Journal of Tropical Insect Science*, 27, 114–121.
<http://dx.doi.org/10.1017/S1742758407814676>
- Kanyamasoro, M. G., Karungi, J., Asea, G., & Gibson, P. (2012). Determination of heterotic groups of maize inbred lines and the inheritance of their resistance to the maize weevil. *African Crop Science Journal*, 20, 99–104.
- Kelvin, J. M. (2002). *Maize kernel components and their roles in maize weevil resistance*. Mexico City: International Center for Improvement of Wheat and Maize (CIMMYT).
- Kim, S., & Kossou, D. (2003). Responses and genetics of maize germplasm resistant to the maize weevil *Sitophilus zeamais* Motschulsky in West Africa. *Journal of Stored Products Research*, 39, 489–505.
[http://dx.doi.org/10.1016/S0022-474X\(02\)00056-5](http://dx.doi.org/10.1016/S0022-474X(02)00056-5)
- Masasa, R. T., Setimela, P. S., & Chiteka, Z. A. (2013). Evaluation of open pollinated varieties of maize for resistance to the maize weevil in a controlled temperature and humidity laboratory in Zimbabwe. *Euphytica*, 193, 293–302.
<http://dx.doi.org/10.1007/s10681-013-0900-8>
- McDonough, M. X., Campabadal, C. A., Mason, L. J., Maier, D. E., Denver, A., & Woloshuk, C. (2011). Ozone application in a modified screw conveyor to treat grain for insect pests, fungal contaminants, and mycotoxins. *Journal of Stored Products Research*, 47, 249–254.
<http://dx.doi.org/10.1016/j.jspr.2011.04.001>
- Mohsenin, N. N. (1980). *Physical properties of plant and animal materials*. New York, NY: Gordon and Breach Science.
- Nesci, A., Barra, P., & Etcheverry, M. (2011). Integrated management of insect vectors of *Aspergillus flavus* in stored maize, using synthetic antioxidants and natural phytochemicals. *Journal of Stored Products Research*, 47, 231–237. <http://dx.doi.org/10.1016/j.jspr.2011.03.003>
- Okiwelu, S. N., Adu, O. O., & Okonkwo, V. N. (1987). The effect of *Sitophilus zeamais* (Mots) (Coleoptera: Curculionidae) on the quality and viability of stored maize in Nigeria. *International Journal of Tropical Insect Science*, 8, 379–384.
<http://dx.doi.org/10.1017/S1742758400005427>
- Pingali, P. L., & Pandey, S. (2001). Meeting world maize needs: Technology opportunities and priorities for the public sector. In P. L. Pingali (Ed.) *CIMMYT 1999–2000. World maize facts and trends* (pp. 1–3). Mexico City: CIMMYT.
- Ruswandi, D., Nono, C., Susanto, A., & Puspita, F. D. (2009). Preliminary study on resistance against maize weevil (*Sitophilus zeamais*) in tropical maize lines. Retrieved from <http://repository.unpad.ac.id/handle/123456789/1205>
- Santos, J. P., Maria, J. D. G., & Cruz, I. (1990). Damage to germination of seed corn caused by maize weevil (*Sitophilus zeamais*) and Angoumois grain moth (*Sitotroga cerealella*). *Pesquisa Agropecuaria Brasileira*, 25, 1687–1692.
- Shiferaw, B., Prasanna, B. M., Hellin, J., & Bänziger, M. (2011). Crops that feed the world 6. Past successes and future challenges to the role played by maize in global food security. *Food Security*, 3, 307–327.
<http://dx.doi.org/10.1007/s12571-011-0140-5>
- Siwale, J., Mbata, K., Mrobert, J., & Lungu, D. (2009). Comparative resistance of improved maize genotypes and land races to maize weevil. *African Crop Science Journal*, 17, 1–16.
- Suleiman, R., Williams, D., Nissen, A., Bern, C. J., & Rosentrater, K. A. (2015). Is flint corn naturally resistant to *Sitophilus zeamais* infestation? *Journal of Stored Products Research*, 60, 19–24.
<http://dx.doi.org/10.1016/j.jspr.2014.10.007>

- Tefera, T., Demissie, G., Mugo, S., & Beyene, Y. (2013). Yield and agronomic performance of maize hybrids resistant to the maize weevil *Sitophilus zeamais* Motschulsky (Coleoptera: Curculionidae). *Crop Protection*, 46, 94–99.
<http://dx.doi.org/10.1016/j.cropro.2012.12.010>
- Tracy, W. F., Chandravadana, P., & Galinat, W. C. (1978). More on pericarp and aleurone thickness in maize and its relatives. *Maize Genetics Cooperation Newsletter*, 52, 60–61.
- Vasal, S. K., Villegas, E., Bjarnason, M., Gelaw, B., & Goertz, P. (1980). Genetic modifiers and breeding strategies in developing hard endosperm *o2* materials. In W. G. Pollmer & R. H. Phipps (Eds.), *Improvement of quality traits of maize grain and silage use* (p. 37). The Hague: Nijhoff.
- Yadav, O. P., Karjagi, C. G., Kumar, B., Jat, S. L., Chawla, J. S., Kaul, J., ... Dhillon, B. S. (2014, October 30–November 01). Maize improvement in India. In B. M. Prasanna, B. K. Vivek, A. R. Sadananda, D. Jeffers, P. H. Zaidi, C. Boeber, ... K. Pixley (Eds.), *12th Asian Maize Conference & Expert consultation on maize for food, feed and nutritional security, book of extended summaries* (pp. 263–273). Bangkok.
- Yan, D., Duermeyer, L., Leoveanu, C., & Nambara, E. (2014). The functions of the endosperm during seed germination. *Plant Cell Physiology*, 55, 1521–1533.
<http://dx.doi.org/10.1093/pcp/pcu089>
- Zakka, U., Lale, N. E. S., & Umeozor, O. C. (2015). Efficacy of combining varietal resistance with harvest time and planting date for the management of *Sitophilus zeamais* Motschulsky infestation in stored maize. *Journal of Stored Products Research*, 60, 31–35.
<http://dx.doi.org/10.1016/j.jspr.2014.10.005>
- Zunjare, R., Hossain, F., Muthusamy, V., Choudhary, M., Kumar, P., Sekhar J. C., ... Gupta, H. S. (2015). Popping quality attributes of popcorn hybrids in relation to weevil (*Sitophilus oryzae*) infestation. *Indian Journal of Genetics and Plant Breeding*, 75, 510–513.
- Zunjare, R., Hossain, F., Muthusamy, V., Jha, S. K., Kumar, P., Sekhar, J. C., ... Gupta, H. S. (2015). Genetics of resistance to stored grain weevil (*Sitophilus oryzae* L.) in maize. *Cogent Food & Agriculture*, 1, 1075934. doi:10.1080/23311932.2015.1075934
- Zunjare, R., Hossain, F., Thirunavukkarasu, N., Muthusamy, V., Jha, S. K., Kumar, P., & Gupta, H. S. (2014). Evaluation of specialty corn inbreds for responses to stored grain weevil (*Sitophilus oryzae* L.) infestation. *Indian Journal of Genetics and Plant Breeding (The)*, 74, 564–567.
<http://dx.doi.org/10.5958/0975-6906.2014.00890.6>



© 2016 ICAR-IARI, New Delhi. Published by Taylor & Francis. This open access article is distributed under a Creative Commons Attribution (CC-BY) 4.0 license.

You are free to:

- Share — copy and redistribute the material in any medium or format
 - Adapt — remix, transform, and build upon the material for any purpose, even commercially.
- The licensor cannot revoke these freedoms as long as you follow the license terms.

Under the following terms:

- Attribution — You must give appropriate credit, provide a link to the license, and indicate if changes were made. You may do so in any reasonable manner, but not in any way that suggests the licensor endorses you or your use.
- No additional restrictions

You may not apply legal terms or technological measures that legally restrict others from doing anything the license permits.



Cogent Food & Agriculture (ISSN: 2331-1932) is published by Cogent OA, part of Taylor & Francis Group.

Publishing with Cogent OA ensures:

- Immediate, universal access to your article on publication
- High visibility and discoverability via the Cogent OA website as well as Taylor & Francis Online
- Download and citation statistics for your article
- Rapid online publication
- Input from, and dialog with, expert editors and editorial boards
- Retention of full copyright of your article
- Guaranteed legacy preservation of your article
- Discounts and waivers for authors in developing regions

Submit your manuscript to a Cogent OA journal at www.CogentOA.com

