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BREEDING POTENTIAL OF PHILIPPINE TRADITIONAL MAIZE VARIETIES

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SUMMARY

Maize in the Philippines has been evolving through natural and human-directed selection for more than 500 years. Forty-eight initially collected Philippine traditional maize varieties or populations were characterized with emphasis on tolerance and/or resistance to biotic and abiotic stresses. The varieties exhibited high variability for 9 qualitative and 15 quantitative traits. Several varieties showed outstanding performance in terms of downy mildew resistance, potential corn borer resistance, lysine content and earliness.

Key words: Asian corn borer resistance, downy mildew resistance, heterotic grouping, lysine content, Philippine traditional maize

Key findings: Characterization and screening of 48 initially collected Philippine traditional maize varieties showed high phenotypic variability and several varieties with outstanding performance in terms downy mildew resistance, potential corn borer resistance, lysine content and earliness.

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INTRODUCTION

Maize (*Zea mays* ssp. *mays*) originated in the Meso-american region. It was introduced into the Philippines through the Manila-Acapulco Galleon trade during the Spanish colonial period (1521-1898). Maize in the Philippines has been cultivated and evolving through natural selection and human-directed plant breeding for more than 500 years. Considerable genetic variability and rich genetic resources are expected to be found in different parts of the country where maize is a big part of the diet, income and culture of the people. The potential of the country's indigenous traditional maize genetic resources remain poorly utilized in private and public plant breeding programs.

Some of the most important problems facing the Philippine maize industry are the Philippine downy mildew disease, Asian corn borer infestation, low protein quality in white maize, and inclement weather conditions (i.e. drought, flooding and typhoons).

Historically, the Philippine corn downy mildew disease causes annual maize yield losses of 40 to 60% across the Philippines (Magil *et al.*, 2006). Disease severity is highest during the wet season. First symptoms appear as chlorotic leaf

stripes as early as 9 days after planting. Spores of causal organism, *Peronosclerospora philippinensis* (Weston) Shaw are produced on the underside of leaves which appears as downy or powdery substance. The disease ultimately causes tassel malformation, stunted ear formation and seed sterility.

Asian corn borer, *Ostrinia furnacalis* (Guenee) is a major insect pest in maize. Damage due to this destructive pest can cause 20 to 80% yield losses (Sanchez, 1971) or 10 to almost 100% yield losses according to farmers (Mutuc *et al.*, 2012). Bt corn definitively reduces yield loss due to corn borer; however, there is still a need to develop open-pollinated cultivars with non-Bt and non-restrictive sources of resistance to this major insect pest.

Lysine is one of the 9 essential amino acids. Quality protein maize (QPM) varieties are nutritionally superior over normal maize. QPM possess higher proportion of lysine and tryptophan than normal maize. Lysine content by weight in normal maize is < 0.40%, while in QPM it is ≥ 0.40%.

Earliness or early maturity in maize is an attractive trait. Generally, current open-pollinated and hybrid maize varieties in the Philippines mature at 120 days after sowing. Early maturing maize varieties are generally preferred because of faster return on investment compared with late maturing varieties. Early maturity is also advantageous when abiotic stresses, i.e. drought, typhoon and flooding, are expected to hit late in the cropping season.

This study aims to characterize 48 Philippine traditional maize varieties or populations with emphasis on resistance to Philippine downy mildew and Asian corn borer, lysine content and earliness. This will be the first in a series of papers on Philippine traditional maize varieties with the ultimate goal of utilizing our traditional genetic resources in plant breeding programs to enhance the Philippine maize industry.

MATERIALS AND METHODS

Genetic materials

Forty-eight traditional maize varieties were collected across the Philippines from 2013 to 2014 (Table 1). These traditional maize varieties were used to screen for Philippine corn downy mildew and Asian corn borer resistance and were characterized in terms of nutritional qualities, earliness and other morphological and physiochemical characteristics.

Downy mildew resistance

The Philippine corn downy mildew pathogen, *Peronosclerospora philippinesis* (Weston) Shaw is an obligate parasite, which cannot be cultured *in vitro*. To ensure availability of downy mildew inocula, spreader rows or downy mildew-susceptible maize plants (IPB VAR6) were planted 2 weeks before planting the test materials every 5 rows of the entries and were infected with downy mildew at V_2 to V_3 vegetative stages.

Test entries were evaluated for downy mildew reaction by exposing them to point source of inoculum or spreader rows. Each entry was planted in a single 5-m row plot spaced 20 cm between hills and 55 cm between rows with 2 replications. IPB VAR6 was used as susceptible check and spreader rows. Disease rating was done 2, 4 and 6 weeks after plant emergence.

The disease rating scale for the evaluation of downy mildew resistance were as follows: resistant (<10% disease incidence; infected plants showed less than 10% leaf area infection), moderately resistant (11-30% disease incidence; % leaf area infection with chlorotic leaf stripes ranged from 10-20%), moderately susceptible (31-50% disease incidence; % leaf area infection ranged from 21-50% with chlorotic lesion covering almost all infected leaves), susceptible (>50% disease incidence; >50% leaf area infection with chlorotic lesion covering almost all infected leaves).

Table 1. Forty-eight traditional native varieties collected from the Philippines.

I.D.	Name	Kernel color	Source
UPLB Cn N1	TiniguibA	white	Unknown
UPLB Cn N2	Basay	white/red	Unknown
UPLB Cn N3	Batik	white	Unknown
UPLB Cn N4	Toledo	white	Cebu
UPLB Cn N5	MemisA	white	Negros Occidental
UPLB Cn N6	Manaka	white/red	Unknown
UPLB Cn N7	Pastilan	white/red	Negros Oriental
UPLB Cn N8	Banlon	white	Bukidnon
UPLB Cn N9	TiniguibB	white	Bukidnon
UPLB Cn N10	CalimpusA	white	Bukidnon
UPLB Cn N11	KabagtikA	white	Bukidnon
UPLB Cn N12	LakhaRedA	white	Bukidnon
UPLB Cn N13	LakhaRedB	white	Bukidnon
UPLB Cn N14	TiniguibC	white/red	Bukidnon
UPLB Cn N15	TiniguibD	white	Bukidnon
UPLB Cn N16	LawaanA	white	Bukidnon
UPLB Cn N17	BulldogA	white	Bukidnon
UPLB Cn N18	SenoritaA	white	Bukidnon
UPLB Cn N19	FarmerVarietyWhite	white	Bukidnon
UPLB Cn N20	FarmerVarietyOrangeA	orange	Bukidnon
UPLB Cn N21	FarmerVarietyOrangeB	orange	Bukidnon
UPLB Cn N22	FarmerVarietyRedA	white/red	Bukidnon
UPLB Cn N23	FarmerVarietyRedB	white/red	Bukidnon
UPLB Cn N24	FarmerVarietyRedC	white/red	Bukidnon
UPLB Cn N25	CalimpusCompostela	white	Compostela Valley
UPLB Cn N26	CotabatoWhite	white	South Cotatbato
UPLB Cn N27	CompostelaWhite	white	Compostela Valley
UPLB Cn N28	BukidnonWR	white/red	Bukidnon
UPLB Cn N29	PoblacionWhite	white	Bukidnon
UPLB Cn N30	TiniguibCompostela	white	Compostela Valley
UPLB Cn N31	BukidnonRed	orange	Bukidnon
UPLB Cn N32	KitaotaoRed	orange	Bukidnon
UPLB Cn N33	ManggahanWhite	white	Bukidnon
UPLB Cn N34	SanJoseWhite	white	Bukidnon
UPLB Cn N35	TiniguibMaramag	white	Bukidnon
UPLB Cn N36	ValenciaOrange	orange	Bukidnon
UPLB Cn N37	SenoritaPangantukan	white	Bukidnon
UPLB Cn N38	RedhorseBukidnon	white/red	Bukidnon
UPLB Cn N39	CalimpusBukidnon	white	Bukidnon
UPLB Cn N40	MusuanWhite	white	Bukidnon
UPLB Cn N41	LawaanBukidnon	white	Bukidnon
UPLB Cn N42	TiniguibCotabato	white	South Cotatbato
UPLB Cn N43	TiniguibQuezon	white	Bukidnon
UPLB Cn N44	SenoritaBusco	white	Bukidnon
UPLB Cn N45	CebuRed	red	Cebu
UPLB Cn N46	Bagabaga	white/red	Negros Occidental
UPLB Cn N47	CalimpusNegros	white/red	Negros Occidental
UPLB Cn N48	AbraGlutinous	white	Abra

Potential corn borer resistance

Three plants from each population at 40 days after planting (DAP) were used in the bioassay. Five leaf discs were cut from each population, using a small metal cylindrical disc cutter with a diameter of 2.3 cm. Each leaf disc was infested with 5 Asian corn borer, *Ostrinia furnacalis* (Guenee) neonates. After 3 days, the surviving larvae were counted and expressed as percent larval survival. At 60 and 75 DAP stalk feeding tunnel length were measured.

Nutritional qualities

Mineral, fat, lysine, and tryptophan contents and antioxidant activity of seeds were determined for each traditional population. Mineral content, expressed as percent crude mineral content was determined by burning the samples in furnace and weighing the resulting ash. Fat content was determined by Soxhlet extraction of lipid from ground maize seed samples. Lysine content, tryptophan content and antioxidant activity were measured using colorimetric method (Tsai *et al.*, 1972 for lysine; Barman and Koshland Jr, 1967 for tryptophan; and Kravic *et al.*, 2009 for antioxidant activity).

Morphological and physiochemical characterization and grouping

The first 48 traditional populations were characterized and grouped based on their morphology and some seed biochemical traits. The following traits were measured: days to tasseling, days to silking, aleurone color (red, purple or colorless), endosperm color (yellow or white), seed texture (flint, dent or glutinous), colors of stem, leaf sheath, spikelet, spikelet base, and anther, total number of leaves, number of leaves above the ear, angle of the ear leaf, and chlorophyll content at 60 DAS.

Heterotic grouping

Two open-pollinated white maize populations: IPB VAR6 and IPB VAR8 were used as testers for the traditional maize populations. The test-cross trials were planted during 2014 wet season in Central Mindanao University, Maramag, Bukidnon. The results were summarized in a table which groups the populations using k-means clustering method (MacQueen, 1967), and by comparing population yield when crossed with IPB VAR6 or IPB VAR8. If the yield of a population is higher when the tester used was IPB VAR6 then the population groups with IPB VAR8 and vice versa.

Statistical Analysis

All statistical analyses were performed in R (R Core Team, 2015). Cluster analysis using both qualitative and quantitative data made use of Gower's dissimilarity measure and unweighted pair group method with arithmetic mean (Gower, 1971 and Anderberg, 1973).

RESULTS AND DISCUSSION

The 48 traditional populations showed high variability for all the traits measured (Figure 1 and Figure 2). The dendrogram (Figure 2) shows 5 groups or clusters with no apparent relationship with geographic origin of the populations. The cluster analysis reveals extensive germplasm exchange across the country.

Three populations showed exceptionally high downy mildew resistance (DMR): UPLB Cn N15 or Tiniguib D (18.8% incidence), UPLB Cn N33 or Manggahan White (25.5% incidence) and UPLB Cn N17 or Bulldog (27.0% incidence). These materials have high potential in developing DMR populations and lines, as well as in improving current open-pollinated populations and lines with high susceptibility to downy mildew.

Two populations exhibited potential resistance to Asian corn borer (ACB) leaf and stem feedings: UPLB Cn N42 or Lawaan Bukidnon (43.3% larval survival on leaves and 21 cm tunnel length in stems) and UPLB Cn N36 or Valencia Orange (49.4% larval survival on leaves and 17.2 cm tunnel length in stems). S_1 families can be extracted from these 2 populations and tested for ACB resistance. Outstanding S_1 families from each population can be recombined for intra-population

improvement and also advanced for inbred line development.

Two populations have exceptionally high lysine contents at par with QPM varieties (≥0.40% of total seed weight): UPLB Cn N34 or San Jose White (0.50% of total seed weight) and UPLB Cn N10 or Calimpus A (0.41%). These 2 populations can be studied further to assess the variability of lysine content within each population, and compare the genetic basis of high lysine with QPM varieties. Yield may be improved while maintaining high lysine content in these populations through population improvement.

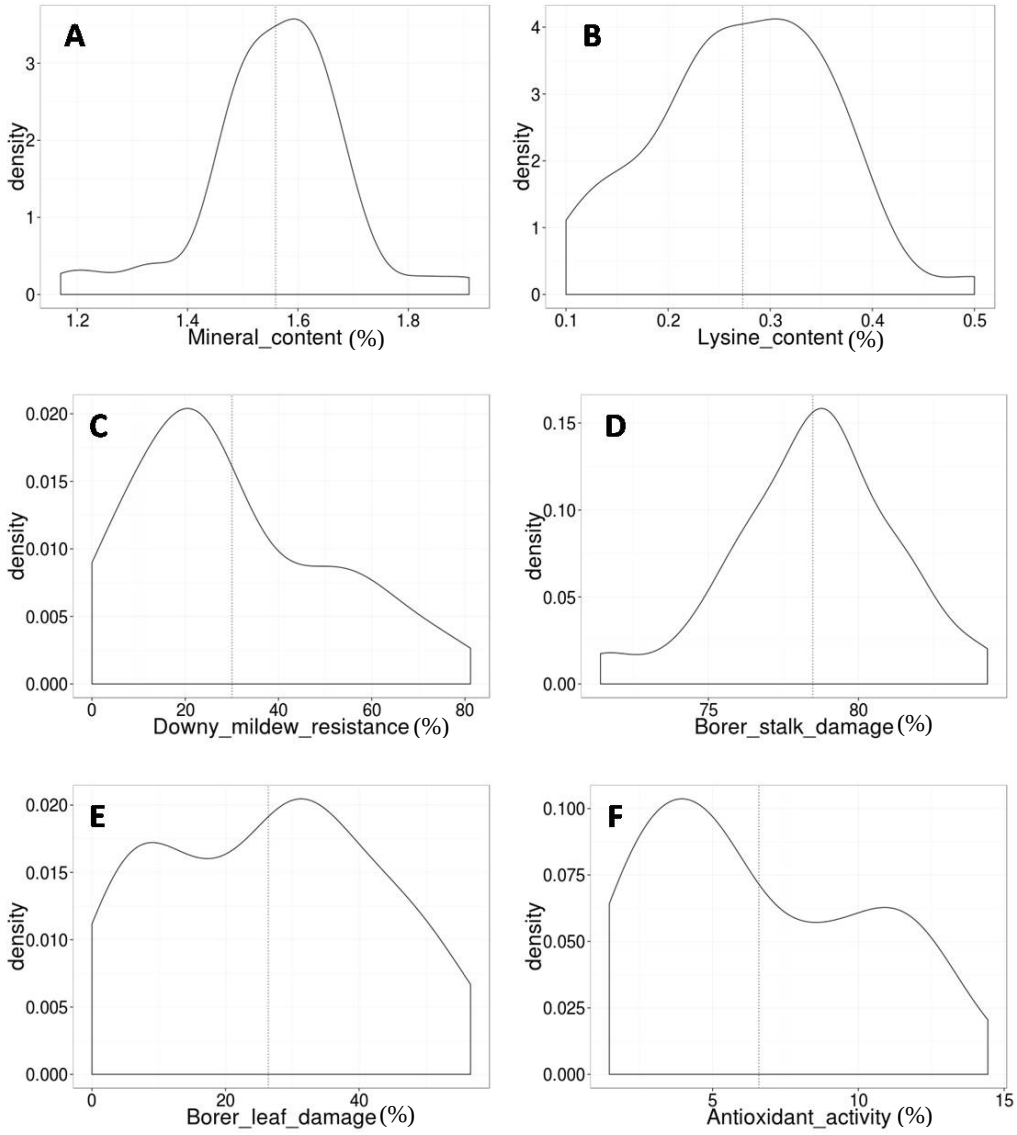

Figure 1. Density plots of mineral content (A), lysine content (B), downy mildew resistance (C), corn borer stalk damage (D), corn borer leaf damage (E) and antioxidant activity (F) for the 48 initially characterized traditional maize varieties.

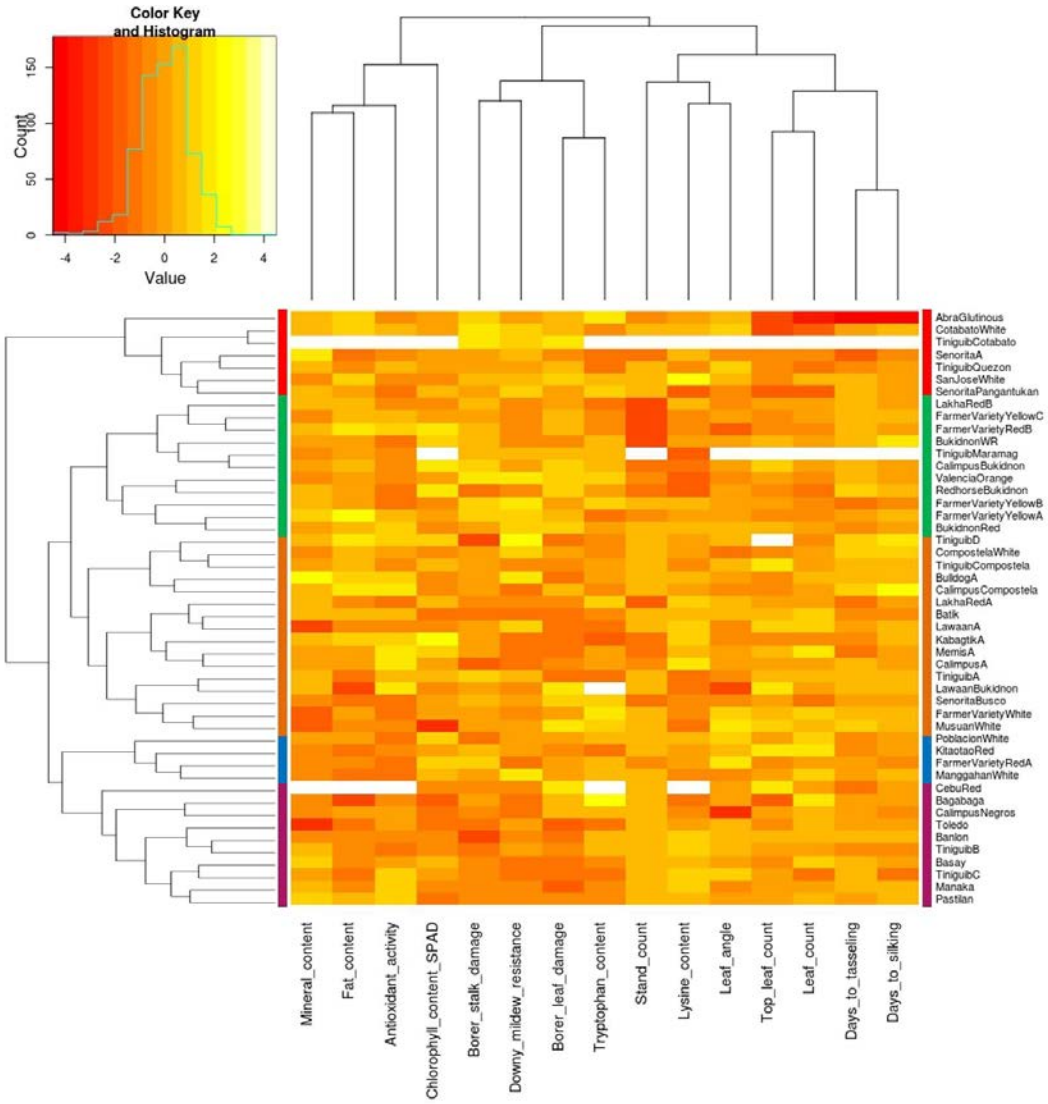

Figure 2. Dendrogram using 24 qualitative and quantitative traits and heat map using the 15 quantitative traits. Dendrogram was cut at 45% dissimilarity and 5 clusters were found.

UPLB Cn N48 or Abra Glutinous is an early maturing traditional maize variety, with anthesis and silking at ~35DAS, green corn maturity at ~55DAS and grain maturity at 90DAS. Yield may be improved while maintaining earliness through population improvement. Inbred lines can also be developed from this population. It can also be used as donor parent for earliness in backcross breeding to shorten cropping duration of current maize varieties.

Test-cross trials using IPB VAR6 and IPB VAR8 as testers are summarized in Table 2. Two heterotic groups were elucidated using k-means clustering and yield comparison when crossed with IPB VAR6 and IPB VAR8. Based on k-means clustering, the first heterotic group consists of generally good combiners with IPB VAR6 (10 populations). The second heterotic group consists of traditional populations that generally combine poorly with IPB VAR6 (24 traditional populations). Members of both heterotic groups can combine well and poorly with IPB VAR8, but the variation in test-cross progeny performance is higher for the first heterotic group.

Table 2. Heterotic groups based on k-means and yield comparison between IPBVAR6 and IPBVAR8 testers.

Traditional population	IPBVAR6	IPBVAR8	Group (K-means)*	Group (yield comparison)**
UPLB Cn N26	1524.40	1429.31	1	1
UPLB Cn N35	1536.10	1884.97	1	2
UPLB Cn N31	1557.32	1572.22	1	2
UPLB Cn N29	1578.22	1457.71	1	1
UPLB Cn N32	1702.10	1361.86	1	1
UPLB Cn N3	1702.82	2497.15	1	2
UPLB Cn N30	1732.20	2291.70	1	2
UPLB Cn N17	2487.16	2279.45	1	1
UPLB Cn N28	2743.50	1522.90	1	1
UPLB Cn N40	2791.76	1908.18	1	1
UPLB Cn N44	253.13	1853.80	2	2
UPLB Cn N19	379.61	1850.49	2	2
UPLB Cn N4	380.00	1575.89	2	2
UPLB Cn N1	381.03	1675.00	2	2
UPLB Cn N8	385.97	2141.22	2	2
UPLB Cn N12	415.20	2144.80	2	2
UPLB Cn N9	445.04	2081.10	2	2
UPLB Cn N5	566.52	1787.73	2	2
UPLB Cn N7	674.72	1283.87	2	2
UPLB Cn N11	675.98	1621.32	2	2
UPLB Cn N18	713.88	2965.20	2	2
UPLB Cn N22	741.24	1601.03	2	2
UPLB Cn N45	747.71	2776.62	2	2
UPLB Cn N42	791.38	910.00	2	2
UPLB Cn N34	795.26	1051.49	2	2
UPLB Cn N24	828.61	1617.62	2	2
UPLB Cn N21	843.18	1913.34	2	2
UPLB Cn N10	893.09	1610.82	2	2
UPLB Cn N15	1029.51	1965.39	2	2
UPLB Cn N38	1042.19	1257.63	2	2
UPLB Cn N2	1043.40	1064.88	2	2
UPLB Cn N6	1062.97	1728.35	2	2
UPLB Cn N25	1078.23	1763.17	2	2
UPLB Cn N23	1082.88	2665.68	2	2
VAR	439,792	231,479	-	-
MIN	253.13	910.00	-	-
MEAN	1039.26	1782.13	-	-
MAX	2791.76	2965.20	-	-

*Group 1 members group together with IPB VAR8 and group 2 members group together with IPB VAR6.

** Grouping based on yield comparison is the result of the conditional statement: if yield when crossed with IPB VAR6 is higher than yield when crossed with IPB VAR8 then the population groups with IPB VAR8, and vice versa.

CONCLUSION

Forty-eight traditional maize varieties or populations were collected from 2013 to 2014 from different provinces across the Philippines. These varieties exhibited high variability for 9 qualitative and 15 quantitative traits. Several varieties showed outstanding performance in terms of downy mildew resistance, potential corn borer resistance, lysine content and earliness: UPLB Cn N15 or Tiniguib D for downy mildew resistance, UPLB Cn N42 or Lawaan Bukidnon for potential corn borer resistance, UPLB Cn N34 or San Jose White for

lysine content, and UPLB Cn N48 or Abra Glutinous for earliness.

There is potential in these Philippine traditional maize varieties to develop open-pollinated and hybrid varieties with improved resistance to downy mildew and corn borer, good nutritional quality in terms of lysine content and earliness. More Philippine traditional maize varieties are currently being evaluated for a wider array of biotic and abiotic stresses. These traditional varieties will ultimately be utilized in breeding programs to develop improved open-pollinated populations, inbred lines and hybrid varieties.

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