

EVALUATION OF DIFFERENT ACCESSIONS / VARIETIES OF MAIZE IN RELATION TO THEIR ANTI-NUTRITIONAL COMPOSITIONS

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ABSTRACT

Maize weevils are known to be among the economically important insect pests of maize in Nigeria. They cause significant damage to maize grains in store. The objective of the study was to assess the susceptibility of different maize accessions / varieties in relation to their anti-nutritional factors. The experiment was done in Department of Crop Science Laboratory, University of Nigeria, Nsukka using a Complete Randomised Design (CRD) and F-LSD to separate the means. Twenty maize accessions / varieties comprising two hybrids (Sammaz – 15 and 16), fifteen open pollinated varieties (Sammaz – 11, 14, 17, 18, 20, 26, 27, 28, 29, 31, 32, 33, 34, 35 and 37) and three local accessions (NSU-P, ENU-E and KAG-W) were used. Sammaz-32 varieties had the least susceptibility index (SI) of 1.79 and was considered as resistant. ENU-E, NSU-P, KAG-W, Sammaz – 11, 14, 15, 16, 17, 20, 26, 27, 28, 29, 31, 33, 34 and 37 were grouped as moderately resistant because their SI ranged between 2.6 and 5.0 while Sammaz – 17, 33 and 35 were grouped as moderately susceptible with an SI range between 5.6 and 7.5. The correlation studies indicated that phytate (0.190), oxalates (0.177), Tannins (0.102), Lectins (0.160) and Trypsin inhibitor (0.106) had very low and positive insignificant ($p < 0.05$) correlations with susceptibility index. However, very low heritability estimated of these qualities suggests that selection of maize accessions / varieties through them will be unreliable. Path coefficient analysis revealed that percentage Tannins (-0.4127) had the highest direct negative on susceptibility index (SI) with a total correlation of (0.102) that was very low and negligibly positive with three opposing positive indirect effects from % phytates (0.3092), %oxalates (0.0235), % lectins (0.2744) and supportive indirect effect from % Try (-0.0899) The heritability estimate of percentage tannins could be considered to make it the most reliable anti-nutritional factor for use in the selection of the maize accessions / varieties in the study for improvement against maize weevil attack.

Keywords: Anti-nutrients, *Sitophilus zeamais*, accessions / varieties, susceptibility index, resistant, correlation studies, path coefficients selection.

INTRODUCTION

Maize, *Zea mays L.* belongs to the family poaceae or gramineae. Maize is an important food and feed in the world, serving as a source of carbohydrates, proteins and minerals (Ogunniyi, 2011). Post harvest storage insect pests cause serious losses to cereals in both quality and quantity and most cases predispose the stored grains to secondary attack by disease causing pathogens (Mwololo, 2013). Dasbak, et al., (2009) reported that developing countries are adapting the use of resistant grain varieties to control grain weevils as a popular alternative to the use of chemicals to avoid attendant risk problems of death, environmental pollution and cost associated with pesticide usage.

Plant species encounter a basic dilemma in the amount of adaptation energy they must allocate to growth versus that, which they must allocate to defenses necessary for survival in the presence of pathogens and herbivores (Chukwulobe, 2013). Plants re faced with a struggle, they must grow fast enough to compete, yet they must also divert energy for the synthesis of secondary metabolites to ward off pathogens and herbivores (Cordain, 2006).

Cereal grains which are the seeds of grasses (gramminae) contain a variety of metabolites which can either be toxic, anti-nutritional, benign or somewhere in-between, depending upon the physiology of the consumer animal (Cordain, 2006). Some secondary

metabolites and their anti-nutritional effects include: Phenolic acid, alkylresorcinols, alpha-amylase inhibitors, lectins, phytates, oxalates, tannins, glycosinolates, cyanogenic glycoside, flavonoids and flavones, etc.

MATERIALS AND METHODS

The study was carried out between April and June 2012, which was the yearly peak period for exponential pest activity of *S. zeamais* in the store within this ecological zone.

The study was conducted at the department of crop science, University of Nigeria, Nsukka, Nigeria. Nsukka is located at latitude 06°52'N and longitude 077°24' and at an elevation of 447.26m above sea level. The prevailing mean temperature and relative humidity during the experimental period were 29±3°C and 75±5%, respectively. The area is characterized with wet and dry seasons. The wet seasons are bi-modal distributed with peaks in July and September of each year. 15 open pollinated varieties (Sammaz – 11, 14, 17, 18, 20, 26, 27, 28, 29, 31, 32, 33, 34 35 and 37), two hybrids (Sammaz – 15 and 16) for the study were gotten from Institute for Agricultural Research (IAR), Ahmadu Bello University, Zaria and three land races (ENU-E, KAG-W and NSU-P). The anti-nutrients (phytates, tannins, lectins, oxalates and chymotrypsin inhibitor in mg/100g maize were determined in IAR food technology laboratory in Collaboration with International Institute of Tropical Agriculture (IITA), Ibadan.

a. Culture of *S. zeamais*

A culture of *S. zeamais* was established to supply similar aged weevils for the experiments. About 10 kg seeds of a white local variety from Kagoro in Kaduna State were procured and cleaned to remove seeds with visible damaged symptoms. The cleaned seeds were fumigated with half phostoxin tablet for two days and later ventilated for two days to free the seeds from the tablet fumes. The seeds were then transferred into four plastic buckets of four litres capacity each and kept in the Crop Science Department Laboratory under environmental conditions of 26±3°C and 65-90%RH for one week. Unsexed adult *S. zeamais* weevils of between 1 and 15 days were collected from infested maize grains in the laboratory and cultured on the cleaned and disinfested maize grains (Kagoro white) in the 4 plastic buckets. Each jar containing 3 kg grains was infested with about 150 weevils. The jars were covered with muslin cloth and fixed with a rubber band to allow aeration and to prevent escape of weevils. Seven days after oviposition, all the weevils were removed from each jar by sieving and the emergent adults (at 36-38 days after oviposition)

considered to be of fairly the same age were harvested and used to infest the maize varieties in the screening test.

b. Screening the maize varieties for susceptibility/resistance to *S. zeamais*

Maize seeds (100g) from each of the maize varieties were placed in 500 ml plastic containers with finely meshed perforations at opposite sides allowing for ventilation and preventing escape of the weevils. The no choice test method, in which the weevils were introduced into each sample of seeds, were as follows. Twenty newly emerged unsexed adult weevils were introduced into the jars to infest the 100 g seeds of each variety and were kept for seven days for oviposition as reported by Abebe, 2009 and Derera *et al.* 2001. Seeds of each variety without *S. zeamais* was kept under similar conditions to served as a control. The treatments were arranged in a Complete Randomized Design (CRD) with three replications on the laboratory platforms for observation at 26-29°C, 65-90% RH and 14:10 (Light: dark) photoperiod.

Index of Susceptibility (SI)

The index of susceptibility was calculated using the method of (Howe, 1971; Dobie, 1974; Gharib, 2004; Dasbak *et al.*, 2009; Abebe, 2009).

SI was determined by finding the log of the total number of emerged adults divided by the median development time of insect from egg to adult multiplied by 100 as follows:

$$\text{Susceptibility Index (SI)} = \frac{\text{Log S}}{T} \times 100$$

Where:

Log S = Logarithm of total number of adult emergents

T = Median Development Time in days.
100 = percentage (%)

The values of susceptibility indices were categorized into five ranks according to Mensah (1986) as follows:

The values between 0.0-2.5 were considered resistant variety (R)

Those between 2.6-5.0 were considered moderately resistant variety (MR)

The values between, 5.1-7.5 will be considered Moderately susceptible variety (MS)

The values between 7.6- 10.0 were considered susceptible variety (S)

Those with SI values greater than 10.0 were considered highly susceptible variety (HS)

Design and Analysis Employed

All the experiments were carried out in the laboratory using Complete Randomised Design (CRD). The data collected were subjected to an analysis of variance procedure and mean differences were compared by the use of F-LSD as outlined by Obi (2002). Correlation matrix was

performed between all the variables assayed. Cause and effect relations of the variables in time and space were determined using path coefficient analysis (Dewey & Lu, 1959) on phytates, oxalates, tannins, lectins and chymotrypsin inhibitor as some of the major causal anti-nutrient components of maize grain susceptibility to *S. zeamais*.

Anti-nutrients composition (mg/100g) of different maize accessions / varieties

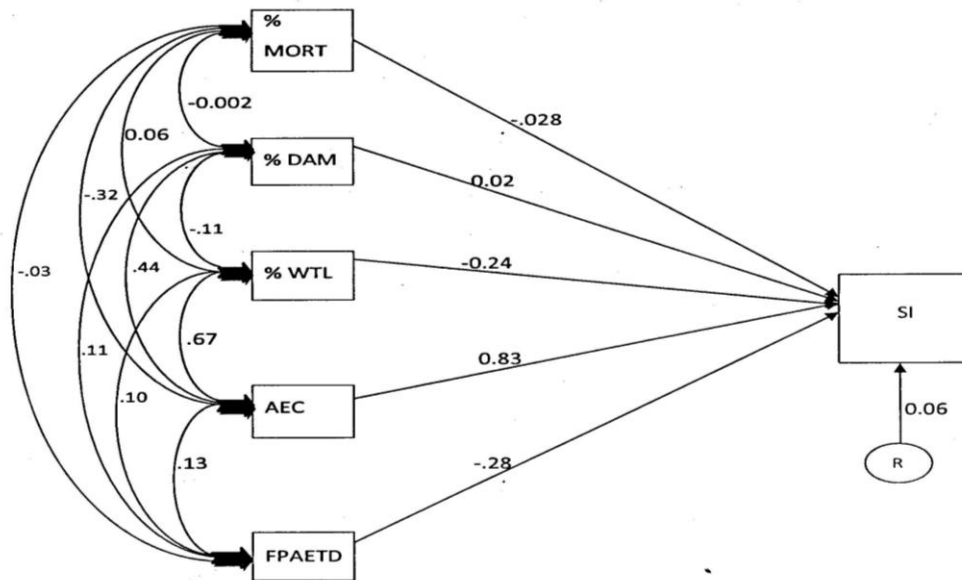
From table 1, it can be seen that each of the 20 maize accessions / varieties contained more tannins than other anti-nutrients. Sammaz 11 had the highest quantity of 0.072 mg/100 of maize followed by Sammaz 15 with 0.058 mg/100g maize both of which remained significantly ($p < 0.05$) higher in phytates than other accessions / varieties. The least quantity of 0.010 mg/100g, maize was recorded in Nsu-p and the general mean for maize accessions / varieties was 0.034 mg phytate per 100 g maize. Sammaz 37 recorded the highest oxalate content of 0.070 mg/100g maize and the least quantity 0.010mg/100g maize was recorded in ENU-E. Sammaz 37 differ significantly ($p < 0.05$) with other accessions / varieties in oxalate content compared with other accessions / varieties. The general mean was 0.038 mg oxalate per 100g maize for each accession / varieties. With respect to tannin content, Sammaz 15 and 29 recorded the highest quantity of 0.480 mg/100g each. Both Sammaz 15 and 29 did not differ significantly ($p > 0.05$) with Sammaz 16, 11 and 18 but they differ significantly ($p < 0.05$) and higher than other maize accessions / varieties. Sammaz 35 recorded the least quantity of 0.120 mg tannin per 100g maize with a general mean of 0.312 mg/100g maize for each accession / varieties. Lectins showed the least concentration in mg/100g maize in all the 21 maize accessions / varieties studied. Sammaz 14, 18 and 29 did not differ significantly ($p > 0.05$) among each other and posses the highest lectin concentration of 0.006 m/100g maize in each. Though Sammaz 14, 18 and 29 also

appeared significantly ($p > 0.05$) similar to Sammaz 20, 28 and 34, they remained significantly different ($p < 0.05$) and higher than other maize accessions / varieties. The least quantity of lectins 0.002 m/100g maize was recorded in ENU-E, NSU-P, Sammaz 31, 32 and 37 which did not differ significantly ($p > 0.05$) among themselves. Sammaz 27 registered the highest concentration of 0.060 mg/100 g maize of trypsin inhibitor which was statistically similar with 0.050 mg/100g maize in Sammaz 20 and 28. Sammaz 37 remained significantly ($p < 0.05$) different from other accessions / varieties but however ENU-E, KAG-W, Sammaz 15, 32, 35 and 37 did not differ significantly ($p > 0.05$) in trypsin inhibitor concentration. The least quantity (0.010 g/100g maize) of trypsin inhibitor was recorded in NSU-P and Sammaz 31 which remained significantly ($p < 0.05$) different and lower than those of other accessions / varieties.

RESULTS

Correlation matrix of susceptibility index of *S.zeamais* INFESTATION ON maize versus anti-nutrients effects

The correlation results in table 2 showed that susceptibility index had though positive but non-significant correlations with phytates ($r = 0.190$), oxalates ($r = 0.177$), Tannins ($r = 0.102$), lectins ($r = 0.160$) and Trypsin inhibitor ($r = 0.106$). A highly significant positive correlation existed between phytates with oxalates ($r = 0.44^{**}$), Tannins ($r = 0.774^{**}$) lectins ($r = 0.589^{**}$) and Trypsin inhibitor ($r = 0.409^{**}$). Oxalates showed a significant positive correlation with Tannins ($r = 0.326^*$) but had highly significant positive correlations with lectins ($r = 0.373^{**}$) and Trypsin inhibitor ($r = 0.477^{**}$). Tannins showed a highly significant positive correlation with lectins ($r = 0.695^{**}$) and Trypsin inhibitor ($r = 0.583^{**}$) while also, a highly significant correlation ($r = 0.774^{**}$) existed between lectins and trypsin inhibitor.



% MORT = Percentage mortality, % DAM = Percentage damage, % WTL = Percentage Weight Loss, AE = Adult emergence, FPAETD= Fifty percentage adult Emergence Time in days, R=Residual

FIGURE 1: Path diagrams and coefficients of *S. zeamais* development and activities in maize to its susceptibility index (SI)

Table 1: Anti-nutrients composition (mg/100g) of different maize accessions / varieties

Variety	Phytates	Oxalates	Tannins	Lectins	Chemotrypsin inhibitor	Susce. Index (si)
Enu-E	0.018	0.010	0.130	0.002	0.020	4.57
KAG-w	0.022	0.020	0.140	0.003	0.020	4.57
NSU-P	0.010	0.020	0.130	0.002	0.010	3.47
SAM-11	0.072	0.040	0.460	0.004	0.040	4.67
SAM-14	0.038	0.040	0.380	0.006	0.040	4.10
SAM-15	0.058	0.040	0.480	0.004	0.020	4.13
SAM-16	0.022	0.020	0.460	0.003	0.040	4.11
SAM-17	0.044	0.060	0.410	0.004	0.040	5.06
SAM-18	0.044	0.040	0.440	0.006	0.040	3.66
SAM-20	0.026	0.040	0.400	0.005	0.050	4.16
SAM-26	0.046	0.040	0.420	0.004	0.040	3.47
SAM-27	0.024	0.060	0.280	0.004	0.060	3.72
SAM-28	0.050	0.050	0.360	0.005	0.050	4.40
SAM-29	0.048	0.040	0.480	0.006	0.040	4.83
SAM-31	0.014	0.020	0.140	0.002	0.010	3.50
SAM-32	0.016	0.020	0.180	0.002	0.020	1.79
SAM-33	0.042	0.050	0.270	0.004	0.040	5.84
SAM-34	0.048	0.040	0.420	0.005	0.030	3.55
SAM-35	0.022	0.030	0.120	0.003	0.020	5.33
SAM-37	0.023	0.070	0.140	0.002	0.020	4.63
MEAN	0.034	0.038	0.312	0.004	0.033	4.03
F.LSD(0.05)	0.0017	0.0017	0.040	0.0017	0.017	0.91
Fpr	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

ENU-E = Enugu Ezike, KAG-W = Kagoro-white, NSU-P = Nsukka-Pink, Sam= Sammaz, F-LSD = Fisher's least significance difference.

Table 2: Correlation matrix of susceptibility index of *S.zeamais* infestation on maize versus anti-nutrients effects

Variables	%PHY	% OXL	%TAN	Variables	%LEC	% TRY	Corr. Coeff.
%PHY	0.3845	0.0323	-0.3318	%LEC	0.2417	-0.0672	0.190
% OXL	0.1708	0.0726	-0.1336	% TRY	0.1484	-0.0823	0.177
% TAN	0.3092	0.0235	-0.4127	Corr. Coeff.	0.2744	-0.0899	0.102
% LEC	0.2526	0.0293	-0.3078		0.3679	-0.1042	0.160
% TRY	0.1720	0.0398	-0.2471		0.2553	-0.1502	0.106
Residual							0.8535

Note: ** Correlation is significant at the 0.01 level

* Correlation is significant at the 0.05 level

n = Number of treatments (20)

Table 3: Direct and indirect effects of the phytochemical contents of maize on the susceptibility index (SI) of *S. zeamais* mot.

	Susceptibility index SI	Phytates	Oxalates	Tannins	Lectins	Trypsin inhibitor
Susceptibility index (SI)	1	.190	.177	.102	.160	.106
Phytates		1	.44**	.774*	.589**	.409**
Oxalates			1	.326*	.373**	.497**
Tannins				1	.695**	.583**
Lectins					1	.774**
Trypsin inhibitor						1

The dependent variable is Susceptibility Index (SI)

Figures in bold prints are the direct effects.

% PHY = Percentage phytates

% OXL= Percentage oxalates

% TA = Percentage Tannins

% LEC = Percentage Lectins

% TRY = Percentage Trypsin Inhibitor

n = Number of treatments (20)

Path coefficient analysis of correlation matrix of susceptibility index of *S.zeamais* infestation on maize versus anti-nutrients effects

The study in table 3 revealed that percentage phytate (0.3845) had a maximum positive direct effect on susceptibility index. It also showed positive indirect effects through percentage lectins (0.2417), percentage oxalates (0.0323) while indirect negative effects were through percentage trypsin inhibitor (-0.0672) and percentage tannins (-0.3318). Percentage lectins (0.3679) also registered a high positive direct effect on susceptibility index, its supportive positive indirect contributions on susceptibility index were through percentage oxalates (0.0293), percentage phytates (0.2526) while the opposing negative indirect effects were through percentage trypsin inhibitor (-0.1042) and percentage tannins (-0.3078). Percentage oxalates (0.0726) showed a positive but negligible direct effect on susceptibility index. Percentage tannins (-0.4127) had a maximum negative direct effect on susceptibility index with a supportive indirect effect from trypsin inhibitor (-0.0899) and opposing indirect effects from % phytates (0.3092), % oxalates (0.0235) and % lectins (0.2744) followed by percentage trypsin inhibitor (-0.1502). The residual factor obtained is 0.85

which indicates that about 15% of the variability in susceptibility index (SI) of maize accessions has been explained.

DISCUSSION

Anti-nutrients are secondary metabolites in plants which help protect them against attack by herbivores. The 20 accessions / varieties of maize used in the experiment contained varied concentrations of phytates, oxalates, tannins, lectins and chymotrypsin inhibitors. From table 1 it can be seen that each of the maize accessions / varieties contained more tannins than other antinutrients. Chukwulobe (2013) reported that the presence of some antinutrients can make nutrients unavailable to the attacking pests because they tie up nutrients, e.g. phytic acid forms insoluble complexes with calcium, zinc, iron and copper.

Falvonoids including tannins chelate metals like iron and zinc thereby reducing their absorption (Beecher, 2003). Chukwulobe, 2013 reported that antinutrients also inhibit digestive enzymes and precipitate protein, thus even when the pests are there, their growth and reproduction will be greatly discouraged. Antinutrients have been reported to play protective roles in plants (Varm and Dubey, 2005) thereby discouraging

attack of such plants or plant parts by insect pests (Potenza et al., 2005). Various research reports have shown that these biochemical contents of plants exert their effects in various ways on target pests including, insecticidal, ovicidal, repellent, growth and developmental inhibition (Jbilou et al., 2006; Mamum et al., 2009 and Hatil, 2009). Although the results in table 2, has shown that the susceptibility index of *S. zeamais* on maize grains exhibited very low or negligible positive correlations with phytates, oxalates, Tannins Lectins and Trypsin inhibitor, the path analysis in table 3 shows that % phytates and % lectins have moderate and positive direct effects on susceptibility index while % Tannins had a moderate and negative direct effect on susceptibility index with a supportive indirect effect through % Try (-0.0899) and opposing positive indirect effects through % TAN (0.3092), % oxl (0.0235) and % LEC (0.2744). These biochemical compounds in the form of phenolic amides such as defeuroyl and dicoumaroyl may be antibiosis factors to the *S. zeamais* (Simbarashe, 2013). Some more recent researchers on resistance to maize weevil in corn grain (Classen et al. 1990, Serratos et al., 1987) have discovered that 90% of phenolic acid compound, mainly in the form of (E) – furulic acid were found concentrated in the pericarp, testa and aleurone layer of the seeds, which play a very important role in suppressing weevil development, most possibly due to antibiosis effect.

CONCLUSIONS

All phytochemicals (anti-nutrients) exhibited moderate to high significant positive correlation with each other and all in turn show negligible non significant correlation with susceptibility index, and so selection of the maize accessions for resistance base on their association values will be unreliable but the path coefficient analysis indicated that % tannins had the highest negative direct effect on *S. zeamais* susceptibility/resistance, indicating a better prospect in the selection of accessions with high tannin concentrations for improvement against *S. zeamais* among the maize accessions.

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