The Journal of Food Technology in Africa Vol. 9 No. 1, 2004, pp. 29-36

Effect of Insecticidal Plant Materials, *Lantana camara* L. and *Tephrosia vogelii* Hook, on the Quality Parameters of Stored Maize Grains

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ABSTRACT
The effect of ground powders of two tropical plants, *Lantana camara* L. and *Tephrosia vogelii* Hook, on the level of insect damage and the grain quality parameters of stored maize were evaluated for five months. The evaluations were aimed at generating natural product treatments suitable for post-harvest grain protection and as sustainable alternatives to synthetic insecticides in the control of the maize weevil, *Sitophilus zeamais* Motschulsky. Three rates (1.0, 2.5 and 5.0% w/w) of each plant powder, a synthetic insecticide, Actellic Super 2% dust at 0.05% w/w and an untreated control were used as treatments. Results showed that the plant powders significantly minimised the magnitude of depression in percent grain moisture content albeit at a lesser rate with high concentration and had no effect on the percent germination of maize grains when compared to the controls. The botanical treatments and synthetic insecticide were equally effective in reducing insect damage by 25%, but the level of damage was independent of the concentration applied. Grain colour and odour were unaffected by the botanicals. Results are discussed with regard to the use of botanicals by small-scale farmers as cost-effective and sustainable alternatives to synthetic insecticides in maize grain storage.

Key words: botanicals, indigenous knowledge, grain storage, grain quality parameters, synthetic insecticides, insect damage, seed viability, food safety

INTRODUCTION

Food security in sub-Saharan Africa largely depends upon improved food productivity through the use of sustainable good agricultural practices (GAPs) and the reduction of postharvest losses caused by pests and diseases. To ensure high food quality and safety standards, which are acceptable to the consumer, quality control, including good storage and handling practices, must be observed at all times (Kenny, 1998; Whitehead, 1998). For decades, the pest control policy in developing countries has been dependent upon the use of synthetic pesticides. Although synthetic pesticides are known to have undoubted benefits, their adoption rate and use for insect pest control in grain storage has remained remarkably low in the resource-poor farming environments. The subsistence nature of agriculture in developing countries coupled with the high cost, poor information and erratic supply of synthetic pesticides have emerged as reasons for farmers' reluctance to adopt pesticides (Tembo and Murfitt, 1995; Ogendo, 2000). Most farmers in these environments store their agricultural produce on the farm for short (less than three months) or long periods (^{3}six months) (Wanjekeche, 1997) and use a variety of indigenous plant species with insecticidal properties for pest control (Bekele *et al.* 1996; Ogendo, 2000). Recent revelations that synthetic insecticides penetrate into stored grain and may be toxic (Lalah and

Wandiga, 1996; El Sheamy *et al.*, 1988), is worrisome. Moreover, Jood *et al.*(1993) reported that plant products and insect infestation adversely affected the taste, aroma and overall acceptability of ?chapatis? from treated maize rendering the grains unsuitable for human consumption. However, oils and crude powders of several plant species have been shown to have no adverse effects on the germination of maize, sorghum, pigeonpea and green gram (Pandey *et al.*, 1986; Kasa and Tadese, 1995; Obeng-Ofori, 1995). This has stimulated scientists to search for natural pesticides that are environmentally friendly, safe to man and other non-target organisms and have no adverse effects on the organoleptic and market quality of stored grains.

Lantana camara L. and *Tephrosia vogelii* Hook have been shown to have toxic and repellent effects against certain insect pests of stored grains (Oliver-Bever, 1986; Sharma *et al.*, 1992; Facknath, 1994; Ogendo, 2000) supporting indigenous farmer practice on their widespread use as grain protectants. However, conclusive recommendations on their use can only be made after the effects of these botanicals on the grain quality and safety are ascertained.

MATERIALS AND METHODS

Test plant materials

Fresh samples (mixture of leaves, inflorescence and succulent stems) of two local plants, *Lantana camara* L. (Wild sage; Verbenaceae) and *Tephrosia vogelii* Hook (Fish poison bean; Fabaceae) were collected from the farm survey areas. The plant samples were dried under shade at room temperature of 27-30ºC for three days and further oven-dried in the laboratory at 35ºC for 48 hours (Wisniewski, 1999). The dry plant samples were ground to fine powder using an electric laboratory hammer mill.

Mass rearing of test insects

Heavily infested maize grains were collected from the survey area and screened in the laboratory for confirmation of the insect species. Two hundred unsexed adult *Sitophilus zeamais* Motschulsky insects were reared in 1- litre jars containing 500g of disinfested whole maize grains as described by Haines (1991). The top of each rearing jar was covered with nylon mesh fastened tightly with rubber bands, and the insects were allowed a 7-day period for egg laying (oviposition). Thereafter, all adults were removed and each jar allowed to stand for 25 days during which emerging adult insects were monitored and kept in separate jars according to their age. The rearing of test insects was done in the laboratory at ambient temperatures of 17-28ºC, 38- 69 % relative humidity (RH) and at L12: D12 regime. A local maize (Zea mays L.) variety, Nyar Maragoli, purchased from Kasipul Division of Rachuonyo District, was used for rearing.

Disinfesting of test maize grains

Maize to be used in the experiments was disinfested under a gas tight sheet using phostoxin tablets at 5 g PH3 per tonne for 10 days to kill any latent insect infestation according to Haines (1991). The disinfested maize was thereafter kept in the laboratory under ambient conditions. Sub-samples used for the grain quality evaluations were further disinfested at 40ºC in an oven for 4 hr (Bekele *et al.*, 1996) and allowed to cool for 2 hr before use.

Grain quality evaluations

Ground powders of *Lantana camara* L. and *Tephrosia vogelii* Hook at three rates (1.0, 2.5 and 5.0% w/w) were admixed with 5 kg of disinfested maize. An untreated sample and a synthetic insecticide treatment, Actellic Super (16g/kg Pirimiphos-methyl + 3g/kg Permethrin) 2% dust at the recommended rate of 0.05% (w/w) were used as comparative controls for the botanical treatments. A total of eight treatments (**Table 1**), each replicated six times, were arranged in a completely randomised design (CRD) on one metre high wooden benches in the laboratory. All treatments were kept at ambient temperature of 17-28ºC, 38-69 %RH and a 12L:12D hr regime. A cylindrical grain sampler (25 mm in diameter) was used to take 250g sub-samples from each replicate according to the methods described by Haines (1991) after 0, 30, 60, 90, 120 and 150 days of grain storage. The sub-samples were used to determine the grain quality parameters: moisture content (%), insect damage (%), seed viability index (%), grain colour and odour.

Grain moisture content

Direct temperature-corrected moisture content (%) readings were recorded for each grain sub-sample using a capacitance grain moisture meter (Eaton®).

Insect damage

The grain sub-samples were assessed for damage arising from natural insect infestations after 0, 30, 60, 90, 120 and 150 days of storage. Each sub-sample was separated into undamaged and insect-damaged grains. The number of grains in each category was counted, weighed and the percentage weight loss (percent insect damage) of maize grains in storage was computed according to the methods described in Haines (1991) as follows:

$$\% \text{ Weight loss} = \frac{UNd - DNu \times 100}{U\,(Nd + Nu)}$$

Where U = weight of undamaged grains,
D = weight of insect-damaged grains,
Nu = number of undamaged grains and
Nd = number of insect-damaged grains.

Seed viability index

The effect of treatments, storage duration and their interactions on seed viability, expressed as the percent germination, was investigated over a 150-day grain storage period. Unbiased sub-samples of 100 undamaged grains were obtained according to the methods described in Haines (1991). The 100- grain sub-samples were germinated on moistened filter paper (Whatman No. 1) in petri dishes arranged in a CRD with six replicates. The experiment was maintained under laboratory conditions described previously. The number of emerged seedlings from each petri dish was counted and recorded after 7 days. The percent germination was computed according to the methods of Zibokere (1994) as follows:

Viability index (%) = (NG*100)/TG

Where NG = number of seeds that germinated and TG = total number (=100) of test seeds in placed in each petri dish.

Grain colour and odour evaluations

The change in colour and odour of untreated and treated maize was assessed on a monthly basis for five months. Grain sub-samples were assessed for change in colour and odour by modifying Kramer's (1956) quick assessment method into a scoring scale of 1-5 defined separately for each of the two parameters. The scores at day 0 represent the values just before grain treatment.

Scoring for change in grain colour was done according to the following scale:

1. No detectable change i.e. natural white with a few yellow grains
2. Slight change (≤5%) from natural white/yellow to light brown
3. Moderate change (>5 to ^{3}30%) from natural white/yellow to brown
4. Great change (>30 to ^{3}50%) from natural white/yellow to dark brown
5. Highly significant change (>50%) making grain unacceptable for human consumption

Scoring for the change in grain odour was done as follows:

1. Grain is odourless
2. Grain has little offensive odour
3. Grain has moderately offensive odour
4. Grain has offensive odour
5. Grain has very offensive odour making grain unacceptable for human consumption

To obtain unbiased scores, each grain sample was coded and presented in a well-lit and ventilated room for assessment. A panel consisting of six independent assessors (Kramer, 1956) scored for change in grain colour and odour. The assessors were allowed into the assessment room, one at a time, in rotation to ensure that their scores were independent from each other. This procedure was repeated on a monthly basis for five months and the same six panellists retained over the entire assessment period (Kramer, 1956). Blank scoring sheets were used for the different assessment dates to ensure that the previous data did not bias subsequent scores.

Experimental design and analysis

Repeated measurements on each variable were obtained from the same experimental units over time. All the data collected were first homogenised using appropriate logarithmic and square root transformations (Gomez and Gomez, 1984) before being subjected to analysis of variance (ANOVA), repeated measures (univariate and multivariate) analysis and means separated using Tukey's studentized (HSD) test (Gomez and Gomez, 1984; Scheiner and Guvevitch, 1993; SAS Institute Inc., 1995). Non-parametric exact tests [Wilcoxon rank sum test (Mann-Whitney U test)] were used to test for any significant changes in grain colour and odour during storage. The choice of this statistical test was based on the manner in which the scoring was done and its power to test the hypotheses under study (Siegel and Castellan, 1988). For purposes of the final presentation of results, the treatment means were converted to the original scale with the transformed means in parentheses.

RESULTS

Grain quality

The results of the baseline grain quality evaluations at the start of the investigation (day 0) are presented in **Table 2**. The effect of storage duration and botanical treatments on the percent grain moisture content (%MC) are presented in **Table 3** and **figures 1a & b**, respectively. There were significant (*P*=0.05) storage duration, treatment and storage duration by treatment interaction effects on the percent grain moisture content. The treatment effects were significant for 60, 90 and 150 days of grain storage. Storage duration influenced the %MC of both untreated and treated grains. The grain MC increased significantly between 0 and 30 days for all treatments including the untreated control. Thereafter, there was a significant reduction (*P*=0.05) in %MC for all the treatments for the next 3 months (up to 120 days) of grain storage. Grains treated with the synthetic insecticide had consistently lower (*P*=0.05) %MC than those treated with botanicals or the untreated control during the entire five months of storage. The magnitude of depression of grain MC was generally less with the higher dosages of the powdered botanical treatments for each plant species. The lowest grain MC was recorded after 120 days of storage. The magnitude of change in MC of grains treated with L. camara and *T. vogelii* powders at 5.0% (w/w) were significantly different from that of the untreated control.

The effect of storage duration and botanical treatments on seed viability is presented in **fig. 2a & b**. Results indicated that the percent germination significantly (*P*=0.05) increased, except for 120 days, with storage duration for both untreated and treated maize grains. Germination was lowest after 120 days of grain storage. However, there was no significant treatment and storage duration by treatment interaction effects on the percent germination. There were no linear and quadratic relationships for either plant species treatment. Correlation analysis results showed that seed viability was weakly correlated with the grain MC, botanical treatments and the storage duration. All treatments, except the synthetic insecticide, recorded a decrease in percent germination for the first 30 days of grain storage. Untreated (control) grains had the lowest viability index which was judged significantly (no overlap of the standard error bars) different from the botanical treatments. Grains treated with Actellic super 2% dust registered increased percent germination during the first 30 days. *L camara* at 5% w/w and the control treatment had the lowest viability index at 120 days compared to its lower dosages (1-2.5% w/w) and Actellic Super. A similar response trend was observed with *T. vogelii* (all dosages) except grains treated with Actellic Super, which were significantly different from the botanical.

The results of the percent insect damage during 150 days of storage of untreated grains and those treated with different rates of *L. camara* and *T. vogelii* powders are presented in **fig. 3**. Results showed significant treatment (P=0.01), storage duration (P=0.01) and treatment by storage duration interaction (P=0.05) effects on the level of insect damage on stored maize grains. The untreated grains suffered significant (P=0.01) percent insect damage compared to grains treated with Actellic Super 2% dust and the botanical treatments. However, there were no significant differences between grains treated with Actellic Super 2% dust and the different rates of the two plant materials tested. Similarly, the concentration of botanical treatments had no linear and quadratic relationships with the level of insect damage recorded.

Figure 4 shows the mean evaluation scores for colour of untreated maize grains and those treated with Actellic Super 2% dust and the three rates each of the ground powders of *L. camara* and *T. vogelii*. Results indicated that there was significant (P=0.01) storage duration, quality assessors, botanical treatments and storage duration by botanical treatment interaction effects on the colour of stored maize grains. Highly significant (P=0.01) differences were recorded among the treatments. The Wilcoxon scores (rank sums) decreased with storage duration but increased with the dosage of the two plant powders applied.

The results of changes in odour during storage of untreated maize grains and those treated with Actellic super 2% dust and three rates each of ground powders of *L. camara* and *T. vogelii* are presented in **fig.5a & b**. Results showed highly significant (P=0.01) storage duration, treatment, storage duration by treatment, treatment by assessors and storage duration by assessors interaction effects on the odour of stored maize grains. The Wilcoxon scores (rank sums) increased with the dosage of ground powder of the two plant materials but decreased with duration of grain storage. The grains treated with Actellic Super 2% dust had the lowest rank scores followed by the untreated grains. Highest rank scores were recorded with *T. vogelii* at 5% w/w compared to *L. camara* at the same dosage. The change in grain odour was strongest early in storage but decreased with storage duration. Throughout the grain storage period, *T. vogelii* powder at 2.5 and 5% w/w imparted the strongest odour followed by *L. camara* powder at the same rates. However, the changes in grain odour, in all the cases, were mainly between odourless and less offensive. Except for *T. vogelii* powder at 5% w/w, all the other treatments had scored less than two on the five-point scale used.

DISCUSSION

Results showed that the moisture content and seed viability of treated and control grains were all significantly influenced by the duration of storage (**Tables 2, 3; Figures 1 a, b & 2 a, b**). Except for the periods 0-30 and 90-120 days, the results showed increased seed viability with duration of storage for all treatments. On the other hand, the effect of botanical concentration was insignificant. This concurs with Kasa and Tadese (1995) who reported that the use of crude powders of 17 botanical plant species for the control of *S. zeamais* on sorghum had no effect on seed germination. Similarly, Pandey *et al.* (1986) reported that petroleum ether extracts of *L. camara* and four other plant species had no adverse effects on the germination of green gram, Vigna radiata. However, storage duration, botanical treatments and their interaction effects influenced the grain MC. An increase in MC during the first 30 days was observed which was negatively correlated to the seed viability index. Thereafter, the grain MC decreased during storage for three months (30-120 days) and the magnitude of depression of MC decreased with concentration of plant powder. The grain MC and viability index were lowest after 120 days of storage. This extremely low MC and viability index observed after 120 days coincided with the time when the ambient relative humidity was only 38%. The changes in grain MC can be attributed to variations in ambient temperature and relative humidity during storage. All durable stored products are hygroscopic and can therefore absorb or desorb (release) moisture from and to the surroundings. The moisture holding capacity of the air increases three times the amount of change in temperature (Haines, 1991). It follows from this argument that stored products release moisture to the surrounding air with a low relative humidity resulting in depressed grain MC. Where the air temperature and relative humidity are variable, the grain MC will also vary. This explanation seems plausible in this study where the ambient relative humidity and temperature were variable throughout the duration of storage. There was an inverse relationship between grain MC and the seed viability index. The seed viability index increased significantly (82 to 97%) for all treatments as the grain MC decreased (14.5 to 13.3%) during the period 30-90 days of grain storage. There was a sharp drop in seed viability index (97 to 78-85%) for all treatments when the grain MC fell below 13.25% between 90 and 120 days. This indicates that 13.3% could be the critical minimum moisture content of stored maize grains below which a marked decrease in seed viability index is realised. Haines (1991) reported that *S. zeamais* thrives best in grains with moisture contents of about 13%.

Statistical analysis showed significant treatment, storage duration and their interaction effects on the percent insect damage on stored maize grains (**Figures 3 & 4**). The level of damage was significantly higher in the untreated (control) grains than in those treated with the synthetic insecticide and different dosages of the ground powders of the two plant materials. However, the differences between the insecticide and plant materials were insignificant. No linear relationships were detected between the concentration of powder applied and the level of insect damage observed. The low moisture content of the grains, extremely dry weather conditions and lack of other grains stored in the neighbourhood could explain the exceptionally low damage levels (between 0.7 and 0.9% for all treatments). These results agree well with Schulten (1996) who worked with Prostephanus truncatus and observed that the progress of infestation in traditional maize varieties in large parts of eastern Africa was very slow during the dry season and that measurable losses only develop after the onset of rains. The level of insect damage recorded (0.7 to 0.9%) fits well into their findings of 0.5-4% during the same months (November-April) of the year. Further research may be needed to establish the seasonality of *S. zeamais* infestation in relation to periods of bumper harvests and crop failures. Also it has been suggested that altitudes above 1500 m are likely to slow down insect breeding and hence cause less damage (Schulten, 1996).

The botanical treatments, storage duration, quality assessors and treatment-storage duration interaction significantly influenced the grain colour and odour evaluation scores (**Figures 5a & 5b**). For both quality parameters, the Wilcoxon scores increased with dosage of plant powder but decreased as storage progressed. This implies that concentration influences the amount of odour emitted by a plant material and that the concentration decreases with time. The decrease could possibly be due to the presence of volatile compounds in the plant tissues, which volatilise easily under ambient conditions. Grains admixed with synthetic insecticide had the lowest rank scores followed by the untreated control, but the differences were insignificant. This observation contradicts the general farmer perception that insecticides impart an offensive odour to maize grain and flour (Ogendo, 2000).

This study would appear to indicate that an insecticide treatment is likely to improve the grain colour and odour; however, further studies are planned to confirm this result. The evaluation results indicated that admixing maize with the botanicals marginally changed grain colour and odour over 5 months of storage and most treatments had mean assessment scores less than two on a scale of five. Jood *et al.* (1993) reported that chapatis (unleavened bread) prepared from sorghum grains treated with plant products were normal in colour, appearance and texture after 6 months but had their taste, aroma and overall acceptability adversely affected by either insect infestation or plant products. This means, therefore, that the colour and odour changes detected were expected to affect the market value of grains marginally.

From the results of these trials, it can be concluded that the use of *L. camara* and *T. vogelii* powders as natural product pesticides in maize storage significantly reduce grain damage with no adverse effects on seed germination. The botanical treatments caused only marginal changes in grain colour and odour, which should have virtually no impact on the local market value of treated grain. Further tests are planned to investigate the penetration of such botanical treatments into stored grain and their potential effects on the organoleptic content of the grain. Studies on the levels of residues remaining on treated food and their potential adverse effects when consumed should be evaluated before the use of these treatments are institutionally promoted as part of a sustainable insect pest management system for farm level storage.

CONCLUSION

It can be concluded that the botanical pesticides and storage duration influence the viability and moisture content of stored maize grains without affecting their colour and odour. There is, therefore, good promise to use the two botanical pesticides as alternatives to the synthetic pesticide, Actellic Super 2% dust.

ACKNOWLEDGEMENTS

This research was part of graduate studies funded by the Association of Commonwealth Universities and the University of Greenwich, UK, Shared Scholarship Scheme. The authors would like to thank the International Centre for Insect Physiology and Ecology (ICIPE), Nairobi, Kenya, for financial support under the DRIP Programme and Dr. A. Odulaja and Ms. Margaret N. Wabiri (Biostatistics Unit, ICIPE) for statistical advise.

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Table 1. Treatments used for the grain quality evaluations

Treatment	Description	Dosage
T1	Untreated control	-
T2	Actellic super 2% dust*	0.05
T3	Ground powder of *L. camara*	1.0
T4	Ground powder of *L. camara*	2.5
T5	Ground powder of *L. camara*	5.0
T6	Ground powder of *T. vogelii*	1.0
T7	Ground powder of *T. vogelii*	2.5
T8	Ground powder of *T. vogelii*	5.0

*Actellic super 2% dust used was a cocktail of an organophosphate, Pirimiphos-methyl, and a synthetic pyrethroid, Permethrin (16g/kg Pirimiphos-methyl + 3g/kg Permethrin).

Table 2. Mean (n= 48) quality characteristics of the test grains (baseline grain samples) at the start (Day 0) of the grain quality evaluation activity

Quality Parameter	Value (±SE where applicable)
Grain moisture content (%)	13.6±0.178
Insect damage (%)	0.00±0.00
Seed viability index (%)	89.4±4.630
Grain Odour	Odourless
Grain Colour	Natural white with few yellow grains

Table 3. Effect of storage duration on the percent grain moisture content

Storage duration (Days)	Mean % MC (n=48)
30	14.38 (2.666)a
60	13.99 (2.638)b
90	14.46 (2.600)c
120	12.89 (2.556)d
150	12.96 (2.562)d
MSD (HSD)*	0.089 (0.0066)

Means of six replicates each of untreated and treated grains over 150 days of storage. Means in a column followed by different letters are significantly different at α=0.05 by Tukey's Studentized range (HSD) test. Figures in parentheses are log-transformed values. MSD (HSD)* is the minimum significant difference at α = 0.05.

Fig. 1a: Change in moisture content of grains treated with *L. camara* powder during storage

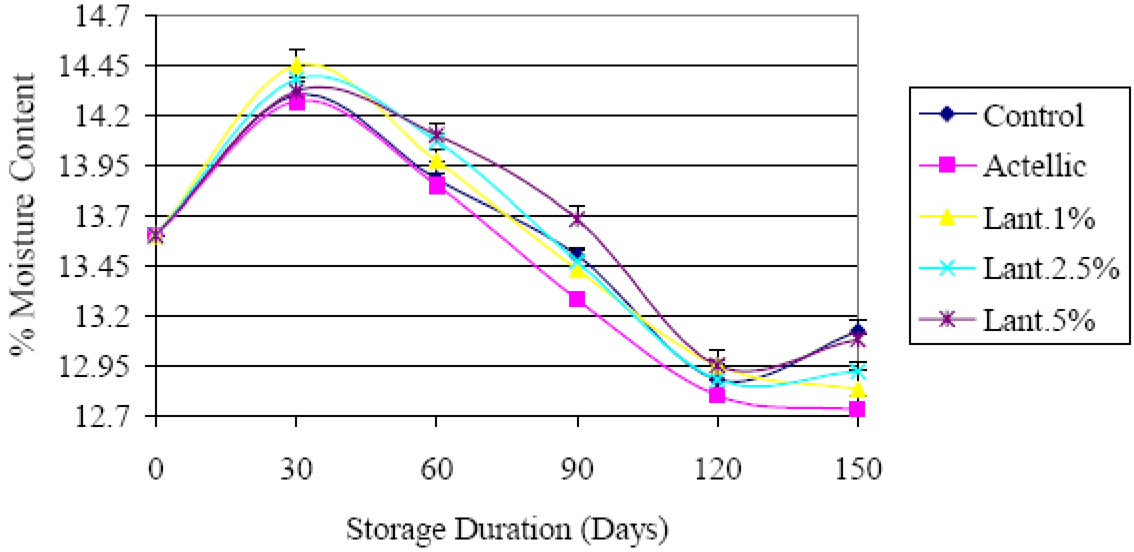

Fig. 1b: Change in moisture content of grains treated with *T. vogelii* powder during storage

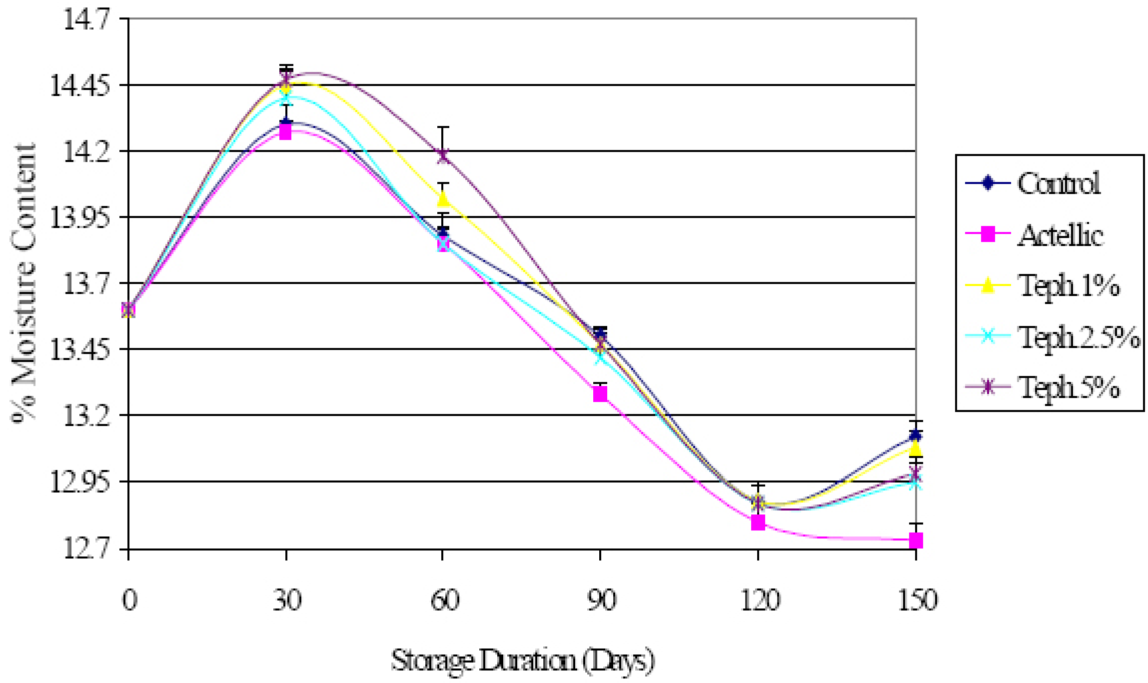

Fig. 2a: Change in viability of grains treated with *L. camara* powder during storage

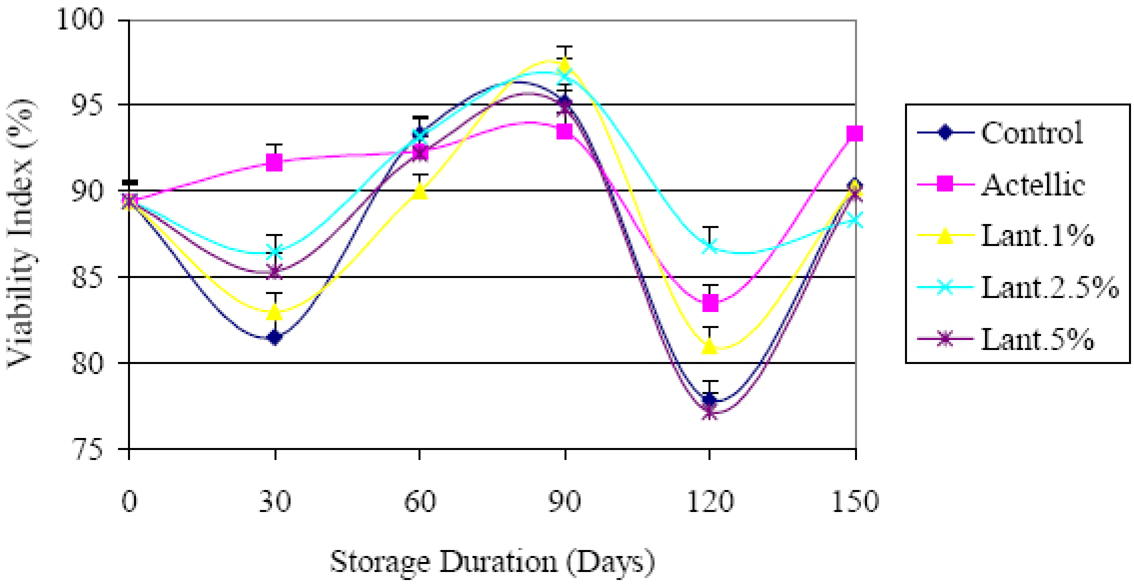

Fig. 2b: Change in viability of grains treated with *T. vogelii* powder during storage

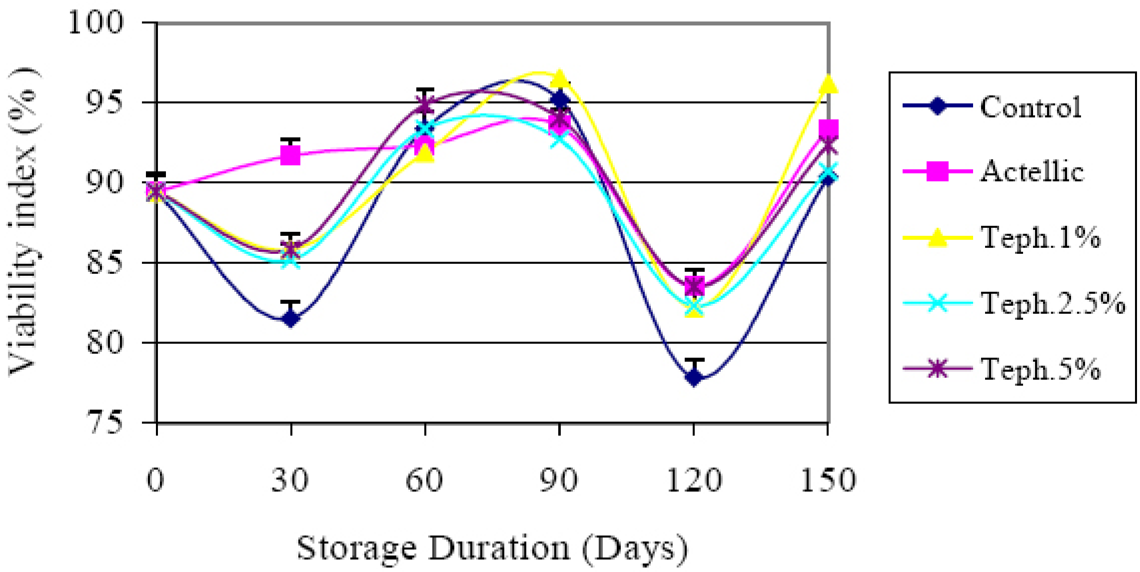

Fig 3 : Effect of *L. camara* and *T. vogelii* powders on percent insect damage of stored maize grains over 5 months

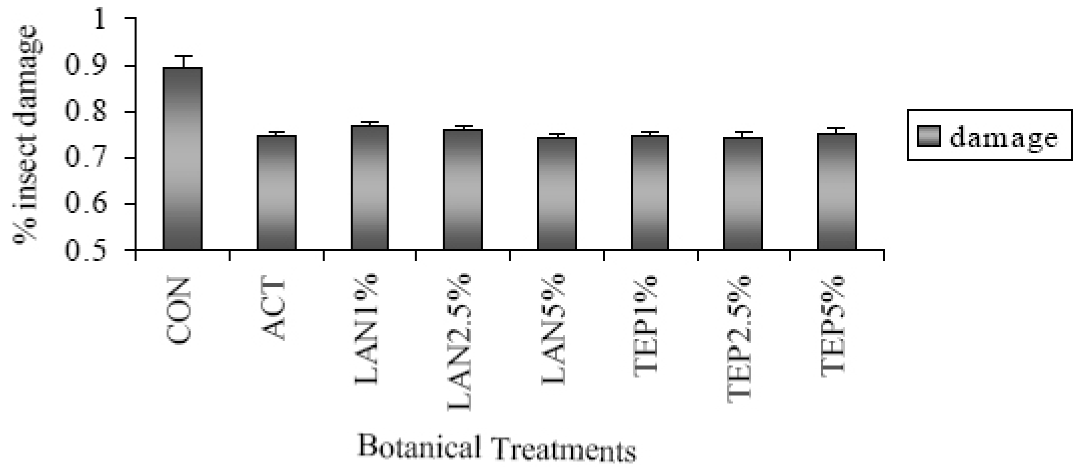

Fig.4: Effect of *L. camara* and *T. vogelii* powders on the mean evaluation scores for grain colour (+SE bars) over a 150-day storage period

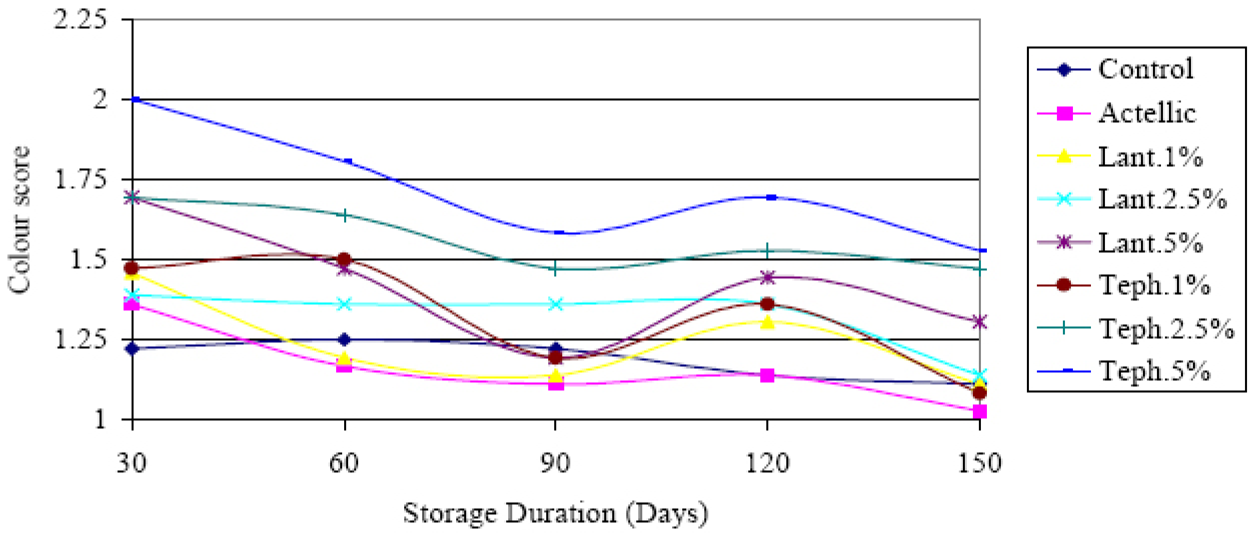

Fig. 5a: Mean evaluation scores for grain odour as affected by storage duration

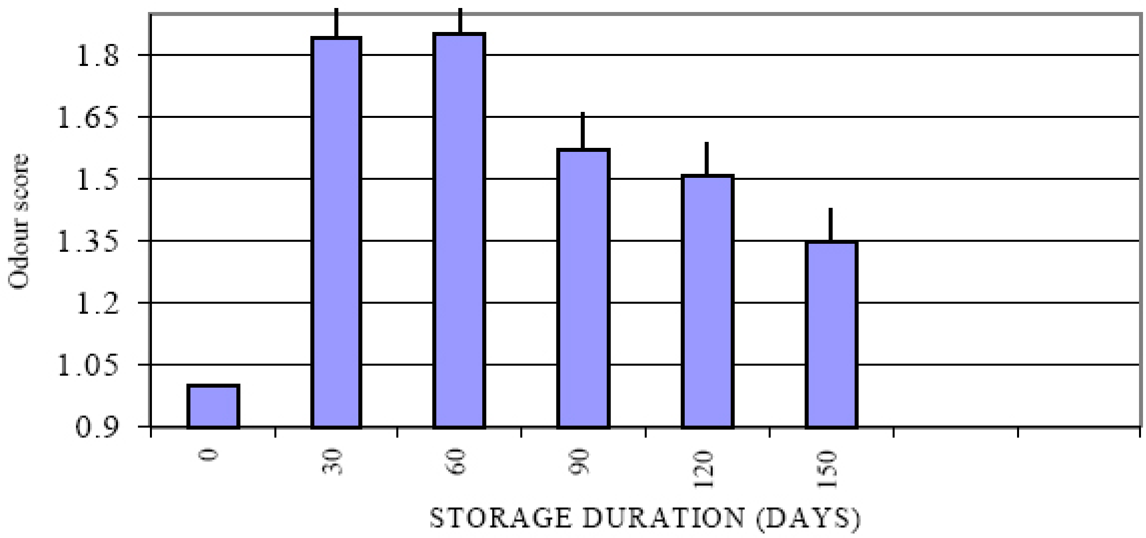

Fig. 5b: Effect of *L. camara* and *T. vogelii* powders on the mean evaluation scores for grain odour during 5 months of storage