Full Length Research Paper

Divergence in technological and diet related parameters of maize treated with essential oils during storage

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Clausena anisata (Rutaceae) and *Plectranthus glandulosus (Lamiaceae)* is applied on the corn grains during conservation limit insect's attacks. In addition to this first use, the present work aims to evaluate the potential of these oils to preserve technological and nutritional qualities of treated grains during 5 months of storage. Investigations carried on grains treated three times consecutively with the effectiveconcentration which kills 80% of experimental population (LC80) of Sitophilus zeamais (Coleoptera: Curculionidae) compared to an industrial molecule imidacloprid, taken as reference. Results show that C. anisata and P. glandulosus essential oils limit grains perforation by S. zeamais during 150 days of storage. In the control sample, S. zeamais bores 83.13% of grains and produces 9.11 g of boring flour after 5 months storage. During the first 50 days, the variation of damages is 19.45, 24.43 and 9.05%, respectively on sample treated with imidacloprid, C. anisata and P. glandulosus. Concerning nutritional parameters following these attacks and treatments, no variation is observed (P>0.05) between samples treated with essential oils and imidacloprid. Principal Component Analysis (PCA) applied on technological and nutritional parameters of food treated with these essential oils shows that, grains treated by P. glandulosus, C. anisata (50 and 100 days) and imidacloprid (50 and 100 days) have a similar level of protection than grains resulting from the control sample stored during 5 months.

Key words: *Clausena anisata*, *Plectranthus glandulosus*, nutritional qualities, technological qualities, *Sitophilus zeamais*.

INTRODUCTION

The quality of corn grains for human consumption depends on three aspects (1) quality of grain for the optimization of grinding, (2) food quality and (3) grain health (CIRAD, 2007). Insects pest destroy these three aspects making the grains unusable. Coleopterans (Arthropoda: Insecta) damage concern quality, quantity, commercial and agronomic value of grains. Most prejudicial species to these stored foods belong to

Sitophilus and *Tribolium* genus (Dal Bello et al., 2001). Generally, their larvae feed preferentially on endosperm or germ, reducing then significantly protein and vitamin food values (Dal Bello et al., 2001). *Sitophilus zeamais* can reduce up to 89.2% of grains in traditional systems of storage during campaign (Nukenine et al., 2002). To conserve the quality of the grain, it is necessary to fight against these devastating insects. Then to preserve the three aspects mentioned above, peasants use chemical insecticides, hazardous for the consumer and the environment (Regnault-Roger et al., 2002). These synthetic insecticides are expensive for the users and
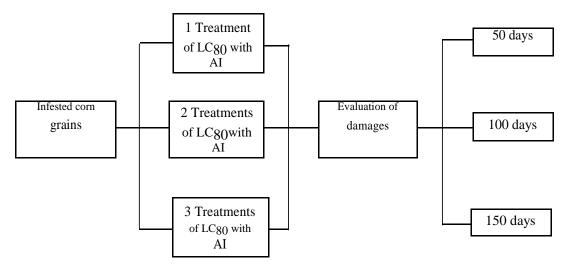
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Figure 1. Diagram of evaluation of technological and nutritional damages of treated corn grains with *C. anisata* and *P. glandulosus* essential oils. AI: Active Ingredient.

may cause potential risk due to the lack of technical knowledge related to their safe use. Among these unexpected effects, the development of resistance by some pests occurred and the lethal effect on non-target species are observed.

It becomes, therefore, useful to build up alternative methods of controlling pest with methods that are user-friendly as the use of agents with high efficacy on the pest and low persistence in the food. There are needs to develop and popularize such control techniques that are clean and friendly as the use of natural essential oils. These natural products as essential oils are often highly specific and biodegradable of low persistence. Essential oils are secondary metabolites abundant in aromatic plants and contain a large number of compounds such as monoterpenes and sesquiterpenes. Essential oils of aromatic plants and spices are tested for their potential as protective agents for human and/or livestock feeds. In this respect thus, to have a protectant edible for human beings or animals, many researches pointed out the use of essential oils from aromatic plants as the best way to control pests without intoxicating human being and animal (Kouninki et al., 2007). Many aromatic plants are known to possess insecticidal effect, to repel ovipositing insects and reduce the progeny (Goudoum et al., 2010). The objective of this study was to evaluate the protective role of *Clausena anisata* and *Plectranthus glandulosus* essential oils on technological and nutritional qualities of treated corn grains after five months of storage.

MATERIALS AND METHODS

Preparation of corn grains samples

The corn grains (CMS 8504) treated three times after 10 days

interval by 500 µl of LC$_{80}$ of essential oils, were transformed into flour using a crusher Polymix (Px-mfc Model, Germany) with mesh of 1 mm.

Determination of the water content of corn grains

The determination of the water content was carried out by Association of Analytical Chemists (AOAC) method (1990) based on measuring sample mass loss after heating at 105°C until complete elimination of interstitial water and the volatile matters.

Evaluation of technological and nutritional damages

It should be noted that, with regard damages, three groups of treated samples were prepared according to Figure 1.

Technological damages

Technological damage returns to physical damages caused by *S. zeamais* on grains. At the end of the conservation time, technological damages were assessed. Technological damages were evaluated on 100 g of corn grains not presenting any crack. The grains treated with LC$_{80}$ of *C. anisata*, *P. glandulosus* and imidacloprid are filled in glass flask (1200 ml) with of 15 couples of *S. zeamais.* The pots were sealed and kept in a dark place for 50, 100 and 150 days. At the end of this period, the number of perforated grains and quantity of boring flour were recorded for each flask. The non infested and untreated grains are considered as a control sample. Four repetitions were carried out. The percentage of perforated grains is expressed according to the formula:

$$\%PGP = \frac{(Tr \times 100)}{Te}$$

Where: Tr= number of perforated grains in treated sample and Te= number of perforated grains in untreated sample.

Evaluation of nutritional parameters on grains

Proportioning of cornstarch in grains

The proportioning of starch was carried out using the method described by Ewers modified (BIPEA, 1978). This method is applicable to cereals and derivatives and comprises of two polarimetric determinations. First, the sample is hydrolyzed hot in hydrochloric medium, and the optical activity (P) measured. Secondly, an alcoholic extract of the sample is hydrolyzed and its optical activity (P') measured. The optical activity (PR) corresponding to starch results from the difference between these two polarimetric measurements.

Calculation and expression of results

Total PR is expressed as a percentage starch compared to the product just as it is; The PR of soluble substances in ethanol is expressed as a percentage starch compared to the dry matter.

$$Starch\,\% = \frac{(P - P').100.100.100}{[\alpha].L.(100 - H°).m}$$

the dry matter at the period of insect attacks is evaluated and removed from the starch value of concerned sample. Hence the quantity of starch contained in boring flour will be expressed by the formula:

$$Y = \frac{(\%A.T)}{100}\,pour\,100\,g\,de\,DM$$

Where: % A: Percentage of starch in infested sample; T: quantity of bore flour according to the dry matter; DM: Dry matter. From these two formulas, one deducts starch quantity contained in infested sample according to the formula:

$$\%\,A(infested) = (1) - (2)\,for\,100\,g\,of\,DM$$

Proteins

The protein content was given according to the Kjeldahl method for the total nitrogen determination (AOAC, 1990).

Evaluation of lipids oxidation state

The evaluation of oxidation state was made according to the Wolf method (1968).

Evaluation of carotenoid loss

The carotenoid loss was highlighted according to AOAC method (1965).

Proportioning of sugars

Sugars were extracted and proportioned by Fischer and Stein

method (1961).

Statistical analysis

The results obtained from the evaluation of technological and nutritional parameters of grains during storage were analyzed with analysis of variance (ANOVA) using the software Statgraphic 5.0. The average values were classified using Duncan Multiple Test with the same software. These technological and nutritional parameters were projected in a system of axis using a multivariate analysis: PCA, with the aim of studying and to visualizing observations between various parameters.

RESULTS

The water content of corn grains

It arises from Figure 2 that the water content is treated with grains samples, those untreated and non infested (control) do not vary. It was located around 84% during 5 months of storage at 28°C and 65 h. No significant difference (P>0.05) was observed between water contents of treated

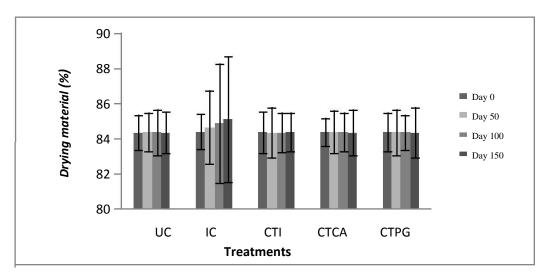**Technological damages on corn grains**

(2)

t arises from Table 1 that technological damages caused by *S. zeamais* evolve with the storage delay. At the LC_{80}, *C. anisata* and *P. glandulosus* essential oils, respectively protect 85 and 95% perforation of grains by *S. zeamais* during 150 days of storage, against 84% with imidacloprid. As regards the quantity of bore flour produced, no significant difference (P>0.05) is noted between these two essential oils and imidacloprid during the storage period. Figure 3 shows the variation rates of grains perforation. At the end of 5 months storage, a *S. zeamais* population at the beginning 15 couples perforates 83.13% of grains and produces 9.11 g of boring flour for 100 g of grains. For samples treated with different active ingredients, damages were most significant during the first 50 days, where a 19.45, 24.43 and 9.05% variation rate were observed, respectively for imidacloprid, *C. anisata* and *P. glandulosus.*

Evaluation of some nutritional parameters on corn grains

The studied nutritional parameters of corn grains obtained from three treatments, control and infested samples are presented in Table 2. These results show a very significant difference (F = 22.15, P<0.01), ndl = (14; 59) between values obtained and analyzed parameters, except for sugars where a significant difference (F = 3.2,
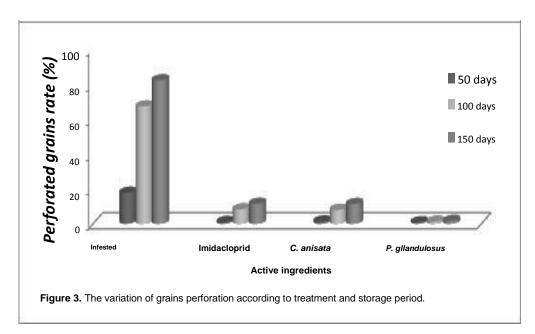
Figure 2. Variation of water content of corn grains according to various treatments applied. UC: untreated corn, IC: infested corn, CTI: corn treated with imidacloprid, CTCA: corn treated with *C. anisata*, CTPG: corn treated with *P. glandulosus.*

Table 1. The reduction rate of perforated grains number and quantity of boring flour according to treatment and storage delay.

Period (days)	Treatment	Perforated grains number	Bore flour quantity
50	Imidacloprid	90.27[b]	97.19[a]
	C. anisata	87.78[b]	98.31[a]
	P. glandulosus	95.48[a]	98.31[a]
100	Imidacloprid	87.95[b]	94.43[a]
	C. anisata	89.76[b]	97.05[a]
	P. glandulosus	96.52[a]	99.18[a]
150	Imidacloprid	84.04[b]	93.96[a]
	C. anisata	84.81[b]	94.73[a]
	P. glandulosus	96.16[a]	99.34[a]

Averages followed by the same letter in the same column are not different significantly with P < 0.05 (Test of Duncan).

Figure 3. The variation of grains perforation according to treatment and storage period.

Table 2. Nutritional variation rate parameters of corn grains according to treatment and storage period.

Period (days)	Treatment	Studied parameters					
		Starch	Protein	Lipid	Carotenoïds	Reducing sugars	Total sugars
50	Infested	-5.61^c	-13.65^b	-22.89^c	-18.51^c	-6.00^b	-8.02^b
	C. anisata	-1.63$_b$	1.62^a	2.39^a	6.95^a	1.32^a	0.57^a
	P. glandulosus	-1.22^b	0.99^a	3.54^a	2.27^a	2.33^a	1.43^a
100	Infested	-5.64^a	-18.75^c	-25.56^c	-56.72^d	-6.76^b	-6.14^b
	C. anisata	-0.19^a	1.82^a	3.26^a	7.29^a	1.99^a	1.72^a
	P. glandulosus	-0.12^a	2.53^a	4.21^a	-0.98^b	2.70^a	2.92^a
150	Infested	-6.81^c	-18.12^c	-31.98^d	-60.00^d	-7.64^b	-6.40^b
	C. anisata	-0.15^a	0.70^a	-6.50^b	-5.00^b	-1.39^a	-1.49^a
	P. glandulosus	0.52^a	0.75^a	1.90^a	-5.63^b	0.69^a	1.16^a

Averages followed by the same letter in the same column are not different significantly with P< 0.05 (Test of Duncan).
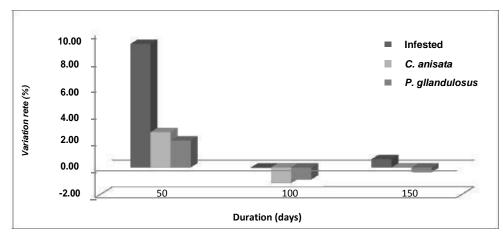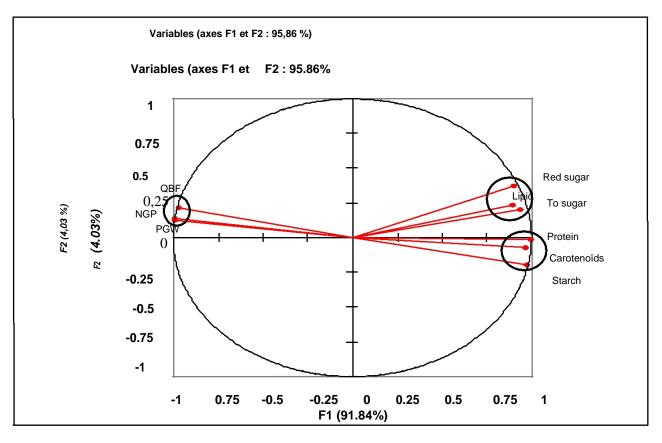

Figure 4. Variation rate of starch in corn grains compared to reference according to storage period.

P<0.05, ndl = (14; 59)) is noted between various treatments. The infested samples show a very significant reduction (P<0.001), respectively after 50 and 150 days of storage of: starch (5.6 to 6.8%), protein (13.6 to 20.8%), lipid (22.9 to 32%), carotenoid (18.5 to 57.6%), and sugars (6 and 8%). The variation between carotenoids quantity is significant between the storage periods and treatments (P<0.001). In the same way, the infestation by insects reduced carotenoid rate during storage. It comes out from analysis of Table 1 that starch variation rate is proportional (r = 0. 97; N = 15) at that of other measured parameters. Figure 4 of starch variation rate, shows that most of damages cause to grains were observable during the first 50 days.
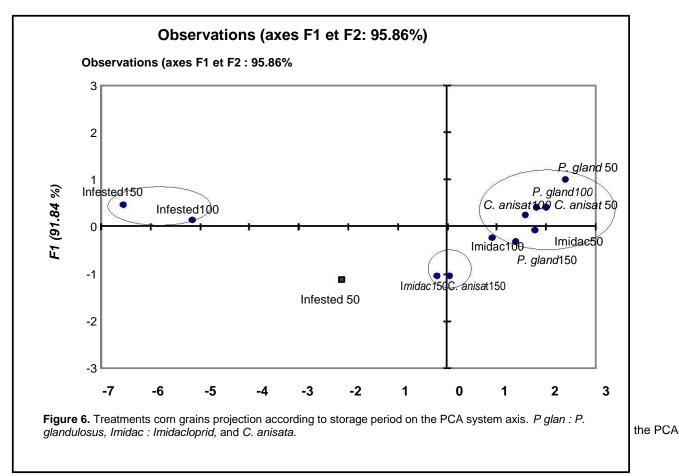
Relation between technological and nutritional damages of *S. zeamais* on grains

Technological properties of grains condition nutritional qualities. These variables attached to a principal component analysis, with the aim of studying and visualizing them, were permitted to present an overall view of similarities and differences between measured parameters (Figure 5). Axes F1 (4%) and F2 (91%) explain results at 95%. The correlation circle (Figure 5) shows and brings together technological and nutritional parameters. The interpretation of correlation matrix shows that all technological parameters are perfectly correlated (R = 1; N = 140) between them, and as well as sugars. Three groups arise from this system of axis: first is represented by the perforated grains number, secondly by the carotenoid rate and protein, and lastly by sugars and lipids oxidation state. When different treatments applied to corn grains are represented in this axis system F1 X F2, they were also gathered in three groups (Figure 6). The first group was represented by the untreated and infested grains storage 100 and 150 days, which translate the number of perforated grains; the second group consists of untreated and infested grains storage

Variables (axes F1 et   F2 : 95.86%)

Figure 5. Projection of studied technological and nutritional coordinate parameters of corn grains on PCA system axis. NGP: Number of perforated grains, QBF: Boring flour quantity, PGW: Perforated grains weight, Red sugar: Reducing sugars, to sugars: Total sugars.

Observations (axes F1 et F2: 95.86%)

Figure 6. Treatments corn grains projection according to storage period on the PCA system axis. *P glan : P. glandulosus, Imidac : Imidacloprid,* and *C. anisata.*

the PCA

system axis.

for 50 days and those treated with imidacloprid and *C. anisata* after 150 days, which translates a weak protein rate and carotenoid; and the last group represented by other treatments: *P. glandulosus* (50, 100 and 150 days), *C. anisata* (50 and 100 days), Imidacloprid (50 and 100 days) and the untreated and safe grains (50, 100 and 150), which translate in addition to other parameters an increase rate of sugar and stable oxidation state.

DISCUSSION

The variation of the water content would be due to the increase of insect metabolic activity consecutive to the increase of their number in the medium (Goudoum, 2010). And the reduction of *S. zeamais* predatory activity is due to insecticidal properties of studied essential oils, which have in their composition bioactive allelochemic molecules (Regnault et al., 2002) which are insecticidal, or antifeeding or both (Ngamo et al., 2007). The starch percentage of treated grains is similar to that obtained by Park et al. (2008) on the millet, but lower than that described by FAO (1993) on the traditional variety. The protein content of corn grains used is far lower than that described by FAO (1995) which is 9%. Beti et al. (1995) showed that, the reduction of oxidation acids fatty rate is mainly due to the metabolic activity of insects, which would have as a consequence the acceleration loss in the unsaturated fatty acids which do not support any temperature rise. This would be explained by the fact that infestation increases the contact surface with oxygen. According to Delia (2001), the availability of oxygen in food is a principal cause of carotenoids oxidation. Taking into consideration this structuring, one can analyze that the treated samples by *P. glandulosus*, *C. anisata* (50 and 100 days) and imidacloprid (50 and 100 days) show a level of conservation close to the control. It is evident and shown by Ngamo et al. (2007) work that treatment with *P. glandulosus* was more effective than that of *C. anisata* and preserves better grains. However, the number of perforated grains is the parameter which varies more in this study conditions.

Conclusion

The results show that the LC_{80} application of *C. anisata* and *P. glandulosus,* respectively reduce at 88 and 98% technological damages and preserve nutritional qualities of grains during the 150 days storage. The critical period of storage is during the first 50 days.

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