

Optimization of Maize Storage Bagged in Triple Bags Containing Aromatic Plants (*Lippia multiflora* and *Hyptis suaveolens*) by Central Composite Design in Côte d'Ivoire

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Abstract: *Lippia multiflora* and *Hyptis suaveolens* are aromatic plants with insecticidal properties. These plants were tested for the efficiency on the stored maize kernels in triple bags. The present study aims to improve, from an experimental design, the methods of maize grain storage in triple bags. A central composite design with five levels represented by three factors affecting the corn storage was used for control the evolution of merchantability (weight losses) and food safety (aflatoxin B1 and activity water) quality during the storage. The factors were: storage time (1 to 18 months), quantity of aromatic plants (0 to 5% of the container mass) and combination of *Lippia multiflora* and *Hyptis suaveolens* (0 to 100% of *L. multiflora*). Results showed that it is possible to assess ideal conditions to keep the maize kernel merchantability quality and food safety during storage. The quality of the kernels maintained for a proportion in aromatic plants greater than or equal to 2.5% during 18 months. In the planned optimal conditions, the experimental values were $3.00 \pm 0.10\%$, $4.81 \pm 0.08 \mu\text{g/kg}$ and 0.78 ± 0.01 for weight losses, aflatoxin B1 and water activity respectively. These values of weight losses, aflatoxin B1 levels and water activity were substantially equal to those predicted by the experimental model.

Keywords: Triple bags, maize, aromatic plants, central composite design, safety quality

1. Introduction

Maize (*Zea mays* L.) is one of the most important cereal crops in the world, both for human consumption and for animal feed. It is the third cereal after wheat and rice with total world production of 1.147.621.938 tons in 2018 [1]. It is made up of approximately 72% starch, 10% protein and 4% fat with an energy value of 365 kCal / 100 g. Corn, compared to cereals, contains more protein and fat. In addition, it contains several vitamins such as thiamin B1, niacin B2, riboflavin B3, vitamin C [2]. To handle the expected current growth of the world population and the seriousness of the problem of food insecurity in Africa, there is a need for increasing of the production and productivity of cereals like maize. However, agricultural production is seasonal while the demand for agricultural products is distributed throughout the year. In this case, the storage of cultures becomes particularly important [3]. Storage is a means by which agricultural or manufactured products are preserved for future use; it is an intermediate phase during the transit of agricultural products from producers to processors and from processors to consumers [4]. Grains must be stored from one harvest to the next to preserve their quality and to maintain their supply throughout the year [5]. Unfortunately, the storage structures used by the farmers are inadequate due to numerous losses due to the presence of pest insects [6]. These insects have a strong impact on the

conservation of corn in the farm environment, i.e. 100% damage after 12 months of storage [7]. Poor storage also leads to mold infestation and mycotoxin contamination [8]. To manage these problems, farmers use chemical pesticides [9]. However, these pesticides have a negative impact on human health and on the environment; hence the search for alternative methods such as the use of biopesticide plants [10]. In recent years, the interest in plant biopesticides has led to the scientific discovery of many aromatic plants including *H. spicigera*, *H. suaveolens*, *L. multiflora* and *L. chevalieri* [11], [12]. *H. suaveolens* and *L. multiflora* are plants found in Côte d'Ivoire and have been the subject of several studies on the postharvest storage of maize and cowpea [13] - [16]. In order to ensure the availability and maintenance of the quality of maize over a long period, a study was carried out on the post-harvest storage of maize in a triple bagging system in the presence of *H. suaveolens* and *L. multiflora*. In addition, this work is to optimize this method from a composite central design.

2. Material and methods

2.1 Site description

The experiment was performed at Research Unit of Biochemistry and Food Sciences (URBSA) UFR Biosciences at the University Felix HOUPHOUËT-

BOIGNY. The different bags were kept in a laboratory storeroom to $27.78 \pm 0.19^\circ\text{C}$ temperature and $75.0 \pm 0.99\%$ relative humidity. Wooden pallets were arranged floored as support for triple bags.

2.2 Collection of maize grains and aromatics plants used in the study

Maize grains and leaves of *L. multiflora* and *H. suaveolens* were collected in March 2016 from producers of Gbêkê Region ($7^\circ 50$ North and $5^\circ 18$ West in center of Côte d'Ivoire). Prior to the storage, maize were sun-dried for 2-3 days before being used for the experiment. While, the *L. multiflora* and *H. suaveolens* leaves were drying at an average temperature of 30°C for 6-7 days and kept away from direct sun exposure. The dried leaves were chopped into fine particles before being used for the experiment.

2.3 Implementation of experiment

2.3.1 Using the Triple Bagging

Storage bags used in our study, were made of polypropylene bags and polyethylene bags (Purdue Improved Cowpea Storage: PICS) developed by Purdue University for storing cowpeas from Niger. These bags, obtained from suppliers, are composed of a triple bagging system.

2.3.2 Protocol of maize storage

The preservation method is based on mixing a proportion of chopped dried leaves and combining the two species studied. The chopped leaves with a quantity of 50 kg of maize kernels were packaged in layers by alternating leaves and corn in triple bags and polypropylene woven bags. The maize was grouped into two categories: the control batches (TPPB0 and TPB0, respectively in the polypropylene bag and the triple bagging) and the experimental batches at different concentrations of *L. multiflora* and *H. suaveolens*.

2.3.3 Central composite design application

A central composite design at five levels (-1.68; -1; 0; 1; 1.68) was used for the realization of storage maize trials. Three independent variables or factors studied were the storage time: from 1 to 18 months (X1), quantity of biopesticides: 0 to 5% w/w (X2) and the combination of *L. multiflora* and *H. suaveolens* (0 – 100 % *L. multiflora*) (X3) (Table 1). The experimental design led to implementation of 17 trials with 8 factorial runs, 8 axial runs (two axial points on each design variable axis at a distance of 1.68 from the design center) and 3 runs at center point. Three experimental

responses were determined. It is about the rate of weight loss, of the concentration in aflatoxin B1 and water activity (Table 2). The coded values of the parameters are replaced by their actual values or states (Table 3) for randomization of the trials. Sampling was carried out at 1, 5, 9.5, 15 and 18 months, in triplicate. Thus, a randomly sample of 2.5 kg from each bag was taken through. At the laboratory the rate of weight losses, the concentration in aflatoxin B1 and the water activity was determined. In the central composite design, the main as well as the interaction effects of various factors are determined by fitting the data into second order polynomial equation:

$$Y_n = b_0 + b_1X_1 + b_2X_2 + b_3X_3 + b_{11}X_1^2 + b_{22}X_2^2 + b_{33}X_3^2 + b_{12}X_1X_2 + b_{13}X_1X_3 + b_{23}X_2X_3 \quad (1)$$

Where Y_n was the measured response, b_0 is the intercept term, b_1 , b_2 and b_3 are linear coefficients, b_{12} is the logarithmic coefficient, b_{11} , b_{22} and b_{33} are quadratic coefficients, and X_1 , X_2 and X_3 were coded independent variables. (Storage time, quantity of plants leaves and plants combination).

2.4 Analysis methods

2.4.1 Assessment of damage and weight loss

To assess the damage caused by insects during storage, samples of 1 kg (approximately 3500 maize kernels) were taken. After sifting and removal of the foreign matters, the grains were weighed and sorted to separate attacked and damaged grains from healthy grains. Then, the two fractions were weighed and counted separately. The percent grain damage was estimated using the method of counting and weighing of [17], [18] described by [19]. Assays were performed in duplicate. Thus, the rate of infection is the ratio of grains having at least one hole in the total number of grains. The estimate of the damage (D) and weight loss (W) is given by the formulas:

$$D (\%) = (NGA / NTG) \times 100$$

NGA = Number of grains attacked; NTG = Total Number of grains

$$W (\%) = [(NGA \times PGS) - (NHG \times WAG)] / (WHG \times NTG) \times 100$$

NGA = Number of grains attacked; NHG = Number of healthy grains; NTG = Total Number of grains; WAG = Weight of attacked grain; WHG = Weight of healthy grains.

Table 1: Independent variables and their coded and actual values used

Independent variables	Coded levels				
	-1.68	-1	0	1	1.68
X1: Storage time (month)	1	4.44	9.5	14.56	18
X2: Proportion of plants leaves (%)	0	1.01	2.5	3.99	5
X3: % of <i>L. multiflora</i> / <i>H. suaveolens</i> (%)	0	20.27	50	79.73	100

Table 2: Code of Matrix for central composite design

Runs	Run Order	Coded values of variables levels		
		X1 (month)	X2	X3 (% Lippia)
Factorials runs	1	-1	-1	-1
	2	1	-1	-1
	3	-1	1	-1
	4	1	1	-1
	5	-1	-1	1
	6	1	-1	1
	7	-1	1	1
	8	1	1	1
Axial runs	9	-1.68	0	0
	10	0	-1.68	0
	11	0	1.68	0
	12	1.68	0	0
	13	0	0	1.68
	14	0	0	-1.68
Center runs	15	0	0	0
	16	0	0	0
	17	0	0	0

Table 3: Experimental values for central composite design

Runs	Run Order	Coded values of variables levels		
		X1 (Month)	X2 (%)	X3 (% Lippia)
Factorials runs	1	4.44	1.01	20.27
	2	14.56	1.01	20.27
	3	4.44	3.99	20.27
	4	14.56	3.99	20.27
	5	4.44	1.01	79.73
	6	14.56	1.01	79.73
	7	4.44	3.99	79.73
	8	14.56	3.99	79.73
Axial runs	9	1	2.5	50
	10	9.5	0	50
	11	9.5	5	50
	12	18	2.5	50
	13	9.5	2.5	100
	14	9.5	2.5	0
Center runs	15	9.5	2.5	50
	16	9.5	2.5	50
	17	9.5	2.5	50

2.4.2 Determination of water activity

The water activity was measured with a HygroLab Rotronic hygrometer according to indications of [20]. Prior to assays, the hygrometer was calibrated with specific water activity salts. Then, samples of 5 g of maize grains were put into standard dry empty containers for the Aw analysis. The water activity digital measures were directly displayed by the hygrometer

2.4.3 Analysis of Aflatoxin

2.4.3.1 Extraction and purification of aflatoxins

Chemical reagents (acetonitrile, methanol and chloroform) and standard aflatoxins (AFB1, AFB2, AFG1 and AFG2) were used for the study. Reagents were purchased from Carlo Erba (Spain) with analytical grade, while standard aflatoxins were provided from Sigma (Sigma, St Louis, MO, USA). Biological aflatoxins (B1, B2, G1 and G2) were extracted and purified from maize using the official guidelines of AOAC [21]. To 25 g of ground maize put in an

erlenmeyer flask, 100 mL of 80% methanol aqueous solution were added. The mixture was homogenized, put in darkness at room temperature for 12 h, and then filtered with a Whatman paper (Whatman N°4). Thereafter, 50 mL of the filtrate were added with 40 mL of a mixture deriving from phosphotungstic acid-zinc sulfate-water (5/15/980, w/w/v), and kept at ambient temperature for 15 min before filtration upon Whatman paper. Aflatoxins were extracted from the out coming filtrate with 3 volumes of 10 mL of chloroform. The extracts were collected into a 50 mL flask and processed with rotative evaporator (BuchiRotavapor R-215) at 40°C for evaporation of the chloroform reagent. Finally, 0.4 mL of hydrochloric acid and 4.6 mL of bidistilled water were added to the dry extract, and the solution was filtered through filter resist in a chromatographic tube then passed through an immunoaffinity column (columnRiDAaflatoxin, Biopharm, Germany).

2.4.3.2 Quantification of Aflatoxins

Determination of aflatoxins contents was achieved with high

performance liquid chromatography column, using a Shimadzu liquid chromatograph (Kyoto, Japan) fitted with fluorescence detector (λ_{exc} 365 nm; λ_{em} 435 nm) and Shim-pack column and pre-column (Shim-pack GVP-ODS: 250 mm x 4,6 mm, 10 x 4,6 mm, respectively). Twenty (20) μ L of the filtrate were injected on the column. Components were eluted with a mobile phase prepared with methanol/water/acetonitrile (60:20:20) and using a gradient programme of 1 mL/min. Assays were performed in triplicate. Validation parameters of the aflatoxins contents analysis, especially Limits of Detection (LOD), Limits of Quantification (LOQ), repeatability and reproducibility traits and percentage of extractions, were valued. Thereafter, the contents of aflatoxins B1, B2, G1 and G2 were estimated, and then the total aflatoxins content was calculated from the sum of the overall aflatoxins. The table 4 presents the HPLC analysis conditions and the results of method validation.

Table 4: Operating conditions of the aflatoxin B1 a proportioning by HPLC and results of the validation method

ITEM	Aflatoxin B1
Pre column	Shim-pack GVP-ODS 10 x 4.6 mm
Column	Shim-pack GVP-ODS, 250mmx4.6 mm
Detector	Fluorescence, λ excitation : 365 nm λ emission : 435 nm
Mobile phase	Acetonitrile/Water/Methanol (20/20/60)
Volume injected	20 μ l
Debit	1 mL/min
Column Temperature	40 °C
Rinsing solvent	Methanol
Analysis time	15 minutes
Limits of detection (LOD)	6.18 ng/kg
Limits of quantification (LOQ)	6.50 ng/kg
Variation coefficient of Repeatability (%)	2.08 \pm 0.10
Variation coefficient of Reproducibility (%)	3.20 \pm 0.18
Extraction yields (%)	98.92 \pm 2. 49

2.5 Statistical Analysis

All experiments were done in triplicate and data in tables and figures represent mean values \pm standard deviation. Multiple linear regression analysis was performed using the Statistica 8 software (Stat Soft, Inc., USA). Experimental data were fitted to the following second-order polynomial model and regression coefficients were obtained. According to the experimental data, the fitting model represented by equation was constructed and the statistical significance of the model terms was examined by regression analysis and analysis of variance (ANOVA).

3. Results

3.1 Experimental responses obtained using Central Composite Design

A central composite design was used to determine the best conditions of maize grain storage in triple bags. The central composite design was developed as presented in the table 5. Weight loss, aflatoxin B1 concentration and water activity were determined. Thus, 17 experiments were conducted

according the matrix presented in Table 5. This table presents also experimental values of weight loss, aflatoxin B1 and the water activity.

3.2 Fitting the models

The various values of the determination coefficients R^2 and R^2 fitted for the regression model of the weight losses; aflatoxin B1 and the water activity were indicated in table 6. These values (respectively of 0.94; 0.98 and 0.97 for R^2) and of (0.86; 0.96 and 0.94 for R^2 adjusted) being roughly close to 1 make it possible to say that the second order polynomial models envisaged, defined well the real behavior of the system. Their non-significant lack of fit also showed that these models were good fit. The lack of fit permitted to justify the adequacy of the model to foresee the variations exactly (Table 6).

3.3 Effects of the variables on the weight losses percentages

The results of the weights losses obtained, while being based on central composite design, are consigned in table 5. Multiple regression analysis was carried on the experimental data and the coefficients of the model are evaluated for the significance. Only, the storage time had significant effects ($P = 0.001$ and $P = 0.05$). The values of the coefficients for the weight losses are presented in table 5. The final predictive equation of the rate of weight loss (Y_1), neglecting the non-significant terms, was given by the equation.

$$Y_1 = 1.15 + 0.68 X_1 - 0.38 X_2$$

The linear terms X_1 and X_2 were significant. These terms had a remarkable impact on the weight losses during storage; while the non-significant terms (X_1^2 , X_2^2 and the interaction between X_1 and X_2 , X_1 and X_3 , X_2 and X_3) have a negligible influence. In order to evaluate the effects of the storage time and quantity of plants on the weight losses of maize during the conservation, Figure 1 is built starting from the equation above. This figure shows the effects of time and the aromatic plants on the rates of weight loss. It indicates that when the variable X_1 is on its higher level and the variable X_2 on its low level, the weight losses increase quickly. The relationships between storage time (X_1) and quantity of plants (X_2) for weight losses are illustrated in figure 1.

As can be seen in the response surface graphs in figure 1, the maximum response values were obtained at high storage time. When the storage time was set at high level, it has observed that the response values were significantly high. While the high plants proportion, it slows down the weight losses.

3.4 Effects of the variables on the aflatoxin B1 contents

Aflatoxin B1 level (AFB1) was affected by storage time and quantity of aromatic plants. The most important parameter affecting level of aflatoxin B1 is the same as in the case of weight losses (factors X_1 and X_2).

$$Y_2 = 1.21 + 1.30 X_1 - 0.19 X_2 + 0.37 X_1^2$$

During storage, linear terms (X_1 and X_2) and quadratic term X_1^2 were significant. On the other hand, the quadratic terms

(X_2^2 and X_3^2) and the interaction between X_1 and X_2 , X_1 and X_3 , X_2 and X_3 are not significant and have a negligible influence on the aflatoxin B1 contents. The surface plot in Figure 2 shows the effect of the time and quantity of plants on the aflatoxin B1 contents. The aflatoxin B1 contents increase significantly in time during the conservation ($P = 0.001$). However, the negative effect of the variable X_2 , starting from a certain concentration threshold, inhibits to a significant degree the aflatoxin B1 concentrations ($P = 0.001$).

3.5 Effects of the variables on the water activity

The multiple regression analysis, executed on the experimental data, permitted to value the coefficients of the

model. These coefficients are evaluated to know the significant effects.

$$Y_3 = 0.74 + 0.02 X_1 - 0.01 X_2$$

All the linear terms (X_1 and X_2) are significant. The significant terms have a remarkable effect on the water activity during the conservation. The storage time and the proportion of aromatic plants have a significant influence ($P = 0.001$ or $P = 0.05$) on the water content. The quadratic terms X_1^2 , X_2^2 and X_3^2 and the interaction between (X_1 and X_2 , X_1 and X_3 , X_2 and X_3) study shows a non-significant influence. Figure 3 indicates the effects of the storage time and the plants proportion on the water activity. Increase in the storage time entails an increase in the water activity and quantity of plants has a negative effect on the increase of the water activity during the conservation.

Table 5: Response surface central composite design and experimental results

Order	Independent variables			Experimental response		
	X1 (Month)	X2 (ratio plants/maize)	X3 (% <i>Lippia / Hyptis</i>)	Y1 (Weight losses %)	Y2 (AFB1 µg/kg)	Y3 (Water activity)
1	-1(5)	-1(1)	-1(20.27)	0.76 ± 0.03	0.14 ± 0.01	0.73 ± 0.01
2	1(15)	-1(1)	-1(20.27)	2.09 ± 0.02	3.05 ± 0.01	0.77 ± 0.01
3	-1(5)	1(4)	-1(20.27)	0.38 ± 0.04	0.04 ± 0.01	0.72 ± 0.01
4	1(15)	1(4)	-1(20.27)	1.36 ± 0.01	2.25 ± 0.02	0.74 ± 0.00
5	-1(5)	-1(1)	1(79.73)	0.78 ± 0.02	0.15 ± 0.01	0.73 ± 0.00
6	1(15)	-1(1)	1(79.73)	2.12 ± 0.02	2.94 ± 0.04	0.77 ± 0.01
7	-1(5)	1(4)	1(79.73)	0.36 ± 0.04	0.04 ± 0.01	0.71 ± 0.01
8	1(15)	1(4)	1(79.73)	1.39 ± 0.02	1.89 ± 0.09	0.74 ± 0.00
9	-1.68(1)	0(2.5)	0(50)	0.24 ± 0.02	0.04 ± 0.00	0.71 ± 0.02
10	0(10)	-1.68(0)	0(50)	2.28 ± 0.08	1.55 ± 0.06	0.76 ± 0.03
11	0(10)	1.68(5)	0(50)	0.50 ± 0.02	1.20 ± 0.05	0.72 ± 0.01
12	1.68(18)	0(2.5)	0(50)	3.19 ± 0.10	4.81 ± 0.00	0.78 ± 0.01
13	0(10)	0(2.5)	1.68(100)	0.93 ± 0.03	0.92 ± 0.08	0.74 ± 0.00
14	0(10)	0(2.5)	-1.68 (0)	0.90 ± 0.02	0.96 ± 0.02	0.74 ± 0.01
15	0(10)	0(2.5)	0(50)	1.24 ± 0.05	1.31 ± 0.07	0.74 ± 0.01
16	0(10)	0(2.5)	0(50)	1.12 ± 0.01	1.10 ± 0.01	0.75 ± 0.01
17	0(10)	0(2.5)	0(50)	1.06 ± 0.01	1.15 ± 0.01	0.74 ± 0.01

Table 6: Regression coefficients of predicted quadratic polynomial models for weight losses, aflatoxin B1 and water activity.

Coefficients	Coefficients estimated		
	Losses	AFB1	Aw
b₀	1.15***	1.21***	0.74***
Linear			
b₁	0.68***	1.30***	0.02***
b₂	-0.38**	-0.19***	-0.01***
b₃	0.02 ^{ns}	-0.04 ^{ns}	-0.001 ^{ns}
Quadratic			
b₁₁	0.13 ^{ns}	0.37***	0.000 ^{ns}
b₂₂	0.05 ^{ns}	0.001 ^{ns}	-0.002 ^{ns}
b₃₃	-0.12 ^{ns}	-0.15 ^{ns}	-0.002 ^{ns}
Interaction			
b₁₂	-0.08 ^{ns}	-0.20 ^{ns}	-0.004 ^{ns}
b₁₃	0.01 ^{ns}	-0.06 ^{ns}	0.001 ^{ns}
b₂₃	-0.01 ^{ns}	-0.03 ^{ns}	-0.001 ^{ns}
R²	0.95	0.98	0.97
R² adjusted	0.88	0.95	0.94
Lack of fit (P-value)	0.08	0.11	0.68

Significant at $P = 0.05$; *Significant at $P = 0.001$; ns: no significant; R^2 : Regression Coefficient, P : probability, AFB1: Aflatoxin B1; Aw: Water activity.

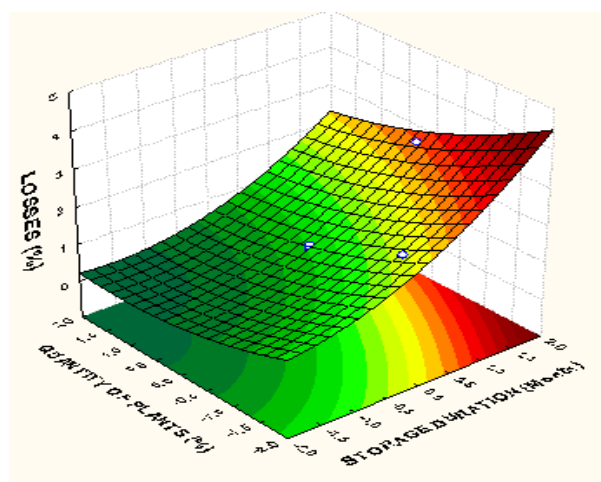


Figure 1: Effects of storage time and quantity of plants on weight losses

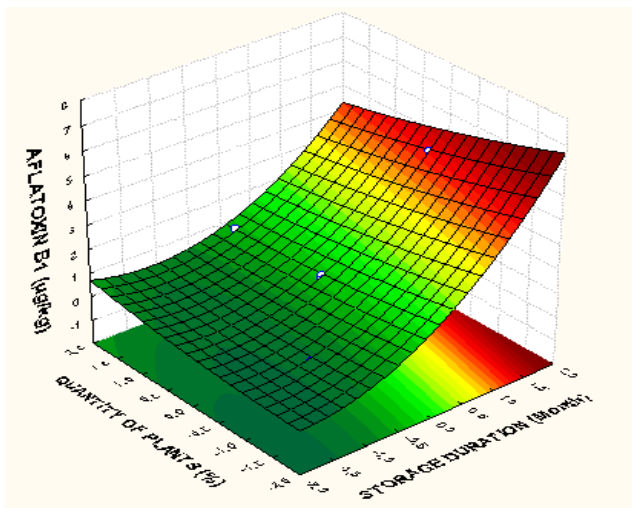


Figure 2: Effects of storage time and quantity of plants on aflatoxin B1 contents

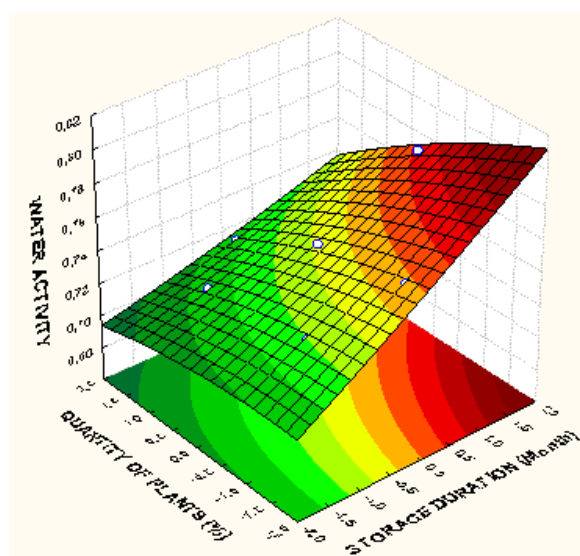


Figure 3: Effects of storage time and quantity of plants on water activity

4. Discussion

The study of the optimization of the storage conditions for corn in a triple bagging system in the presence of plants leaves has shown that there are parameters that significantly influence the marketability and health of stored corn kernels. These parameters are the duration of storage and the proportion of plants leaves. Indeed, losses, aflatoxin concentrations and water activity increase with the duration of storage. The positive sign of the linear coefficient b1 of the variable X1 "Storage duration" for all responses (weight losses, water activity, aflatoxin B1 contents) indicates that the increase in this factor leads to the increase in these responses [22]. Tefera *et al.* [23] have shown that the influence of storage time on losses. According to these authors, the effect of storage time could be explained by the development and activity of insect pests. Regarding the effect of the proportion of aromatic plants on the responses (losses, Aw, AFB1), the linear coefficient b2 is negative. The negative sign of the coefficient indicates that plants of *L. multiflora* and *H. suaveolens* have an inhibitory action on

the development of these responses. Our results are in agreement with those of these authors [13] - [16]. These authors, in their work, have also shown the influence of the duration of storage and the concentration. In addition, this inhibition is observed from a threshold quantity of plants leaves [13]. In our study, a minimum proportion of 2.5% of leaves of *L. multiflora* and / or *H. suaveolens* is sufficient to guarantee the marketable and sanitary quality of the corn kernels. The results presented in this study show that the methods of postharvest of maize storage with the two local species plants, *Lippia multiflora* and *Hyptis suaveolens* in triple bagging system are able to reduce development of pest and alteration of maize. Indeed, lower levels of weight losses, aflatoxin B1 (AFB1) and water activity (Aw) were observed in batches with plants leaves in triple bags during storage. The insecticidal and/or repellent activity of the leaves of these plants would be due to the release of bioactive molecules in their essential oils [24], [25]. According to Tia [26], these insecticidal properties are attributed to the presence of terpenes, such as linalool for *L. multiflora* and β -caryophyllene for *H. suaveolens*. After eighteen months of storage, in triple bags without plants, the concentrations of aflatoxin B1 ($7.33 \pm 0.05 \mu\text{g/kg}$) are superior to normative values [27] and water activity is conducive to aflatoxin B1 increase. By cons, maize batches in triple bags with plants leaves, the aflatoxin B1 values increase only slightly remaining substandard. However it should be noted that a minimum concentration required for optimum efficiency. Tatsadjieu *et al.* [28] showed that the essential oil of *Lippia rugosa*, a species of the genus *Lippia*, inhibits the growth of *Aspergillus flavus* and limit the production of aflatoxin B1 at concentration of 1000 mg/L. Conti *et al.* [29] showed that essential oils of *H. suaveolens* and *H. spicigera* had an effective insecticidal activity. The complete kill of *S. granarius* was observed 24 h after treatment at a minimum effective dose of 0.4 and 0.6 μl per insect with *H. suaveolens* and *H. spicigera* oil, respectively.

The results of the experimental analysis show that conservation is favored when the maize variable storage time is at its highest level (+1) corresponding to 18 months and when the encoded value of the variable amount of plants leaves is at level (0) corresponding to 2.5% of leaves. Thus, the optimum process of maize storage in triple bags containing *L. multiflora* and/or *H. suaveolens* involves the following parameters:

- Storage time: 18 months
- Quantity of plants leaves for storage: 2.5%

By using Statistica 8.0 software desirability function, the ideal conditions of corn kernels conservations were envisaged, with 2.5% of aromatic plants for 18 months. Higher possible values of weight loss and health quality (weight losses, AFB1, and Aw) were determined in table 7. Experimented data were approaching the predicted values in the optimal conditions mentioned above (table 7). This means that there is a high degree suitable between the values observed in the experiment and those predicted by the regression model. For all the parameters of marketable and sanitary qualities of this study, the experimentally obtained values are significantly lower than those obtained in the

control polypropylene bag (TPB0) after 9.5 months of storage (Table 7) and values obtained in triple bags without aromatics plants after 18 months.

5. Conclusion

The results of this study indicate that triple bags in presence of *L. multiflora* and / or *H. suaveolens* leaves are effective for post-harvest maize storage against the insect pests and fungal contamination. This method extends more storage time of maize in Côte d'Ivoire. The use of experimental design showed that it is possible to store maize over a period

of eighteen months without altering the market and healthy qualities of the grain. This study allowed determining the ideal conditions of storage from central composite design. The experimental design has identified optimal storage conditions of maize, it is 2.5% as the minimum concentration of *L. multiflora* and / or *H. suaveolens* leaves for a period of 18 months. This technique is inexpensive, easily carried and fits into the millennium guidelines of respect for the environment. However, this study should be deepened in order to preserve food safety, the nutritional and sensory qualities after storage.

Table 7: Predicted and experimental values of responses under ideal conservation conditions

Responses	Optimal conditions (Triple bags with aromatic plants)		Polypropylene bag without aromatic plants	Triple bag without plants
	Predicted Values	Obtained Values	TPPB0 (9.5 months)	TPB0 (18 months)
Weight losses (%)	2.67 ^b	3.00 ± 0.10 ^b	35.19 ± 0.53 ^a	8.65 ± 0.08 ^a
AFB1 (µg/kg)	4.45 ^b	4.81 ± 0.08 ^b	11.32 ± 0.60 ^a	7.33 ± 0.05 ^a
Aw	0.77 ^b	0.78 ± 0.01 ^b	0.90 ± 0.02 ^a	0.85 ± 0.01 ^a

AFB1: Aflatoxin B1; Aw: Water activity

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