

## Quality of stored grain of corn in different conditions

### Qualidade de grãos de milho armazenados em diferentes condições

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#### Abstract

The growing need to improve the quality of agricultural products and industrial encourages the development of research, aiming at obtaining new methods and techniques that enable the maintenance and preservation of products in storage. The aim of this study was to evaluate the quality of corn and deteriorated quality in two different storage conditions, with low temperatures and high relative humidities (10 °C and 90 % RH) and high temperatures and low relative humidities (30 °C and 40 % RH), featuring two producing regions of Brazil, a part of the Brazilian cerrado and a large part of southern Brazil, during the winter. The experiment was conducted at Federal University of Viçosa, MG, Brazil. It was observed in the results that the storage of corn grains in regions with warm and dry air reduce the risk of incidence of fungi, regardless of the initial quality of the stored product. The incidence of insect was greater for the same conditions. Regardless of storage conditions, bacterial growth was directly related to the quality of the initial product and the storage time. Over time increased risk of aflatoxins and fumonisins, reaching levels above acceptable limits for the quality of silage corn. It was concluded that to maintain the nutritional quality of corn under conditions of 30 °C and 40% of RH is recommended to store it at most six months, whereas in regions with average temperatures of 10 °C and 90% RH are not recommended storing corn for a period exceeding three months.

**Additional keywords:** climates; quality; regions; temperature.

#### Resumo

A crescente necessidade de melhorar a qualidade dos produtos agrícolas e industrializados estimula o desenvolvimento de pesquisas, visando à obtenção de novos métodos e técnicas que possibilitem a manutenção e preservação dos produtos na armazenagem. O objetivo deste estudo foi avaliar a qualidade de grãos de milho deteriorado e de qualidade em duas diferentes condições de armazenamento, com temperaturas baixas e umidades relativas altas (10 °C e 90%) e temperaturas altas e umidades relativas baixas (30 °C e 40%), caracterizando duas regiões produtoras do Brasil, uma parte do cerrado brasileiro e uma grande parte do sul do Brasil, durante o inverno. O experimento foi conduzido na Universidade Federal de Viçosa, MG, Brasil. Observou-se nos resultados que o armazenamento de grãos de milho em regiões de climas quentes e ar seco diminuem o risco de incidência de fungos, independente da qualidade inicial do produto armazenado. Por outro lado, a incidência de insetos foi maior para as mesmas condições. Independente da condição de armazenamento, o desenvolvimento de bactérias foi diretamente relacionado à qualidade do produto inicial e ao tempo de armazenamento. Ao longo do tempo aumentaram o risco da produção de aflatoxinas e fumonisinas, atingindo níveis acima dos aceitáveis para a qualidade do milho armazenado. Concluiu-se que para manter a qualidade nutricional do milho nas condições de 30 °C e 40% de UR do ar, recomenda-se armazená-lo no máximo até seis meses, enquanto que, em regiões com temperaturas médias de 10 °C e 90% de UR não são recomendados o armazenamento do milho por um período superior a três meses.

**Palavras-chave adicionais:** climas; qualidade; regiões; temperatura.

## Introduction

Traditional methods used for quality control, chemical and physical need to be improved, given that it is increasing the demand for better quality products to the consuming public. The control of mycotoxins is of utmost importance, and the best form of prevention is to control the growth of fungi in grains, are extremely important to know all the factors that influence the growth of microorganisms, so that they can be controlled through the steps production, harvesting, drying and storage. Corn is the most widely used raw material in poultry diets (as energy source) according to LIMA (2004), depending on their energy value present in the endosperm. In order to obtain satisfactory quality corn is recommended that the levels of water harvesting and storage, without any risk of deterioration, are 24-32 % (w.b.) at harvest, 13 to 14% (w.b.) up to one year storage and 12% (w.b.) for storage of over one year (WHEINBERG et al., 2008). The temperature and moisture content are the major inducers of fungal growth in the grain (FARONI et al., 2009; CORADI et al., 2011).

Several studies on stored grain indicate that the temperature and relative humidity of the grains mass are the main controlling factors for safe storage (FARONI et al., 2009; CORADI et al., 2011; COSTA et al., 2013). Due to the geographical position of Brazil and its large land area has large climatic variations that can directly affect the quality of corn over a storage period. Between the two regions with the highest grain yield of corn in Brazil (Brazilian savanna and southern Brazil) presented with extreme conditions and different temperatures and relative humidities of the air for the first six months of storage and can be decisive for the product quality.

Thus, it is estimated that to ensure the quality of stored corn is necessary to know the ambient air conditions and temperatures in the storage place for the proper use of technologies for the prevention of quality, as the technique of aeration and cooling. The aim of this study was to evaluate the quality of corn and deteriorated quality in two different storage conditions, with low temperatures and high relative humidities (10 °C and 90%) and high temperatures and low relative humidities (30 °C and 40%), featuring two producing regions of Brazil, a part of the Brazilian cerrado and a large part of southern Brazil, during the winter. The storage of corn was conducted over twelve months and the analysis of grain quality was done every three months.

## Materials and methods

### Experimental design

The grains of corn (*Zea mays* L.) were

stored in sacks in chambers with controlled temperatures and relative humidities of 10 °C and 90%, 30% and 40 °C. A total of 4000 kg of corn used, divided into four batches of 1000 kg. During the storage of corn were monitored temperature and relative humidity of the indoor air daily. Corn samples were collected from 200 grams in all bags, with the aid of a manual auger. The collected product was homogenized, and then removed a simple sample of work for quality assessment. The experiment was conducted in a completely randomized design in a factorial design (2x2x5), two types of corn (quality and deteriorated), two temperatures and air relative humidity storage (10% and 90 °C, 30 °C and 40%) five times of storage (zero, three, six, nine and twelve months). Statistical evaluation of the data was performed using analysis of variance and regression; in case of significant effect of treatment means were compared by Tukey test at 5% probability, using the statistical program Sisvar® 4.3.

### Analysis of quality of the grains corn stored

Corn classification was performed according to the criteria described in the BRASIL (1996). The water content of corn (% w.b.) was determined using the indirect method, the meter of moisture Geole (G-800) after that equipment being calibrated with the official method from the oven, set at 103 °C ± 2 °C for 24 h (BRASIL, 1996). The assessment of insects in the products was made by visual counting in 1 kg of product sampled (LORINI et al., 2002). Identification keys are used for species-level identification of insects (PEREIRA & SALVADORI, 2006). Germination and accelerated aging of corn was conducted according to the Rules for Testing Seeds (BRASIL, 1996). The physicochemical quality (gross fiber, crude protein, ethereal extract, gross energy, dry matter, not extract nitrogen, copper, manganese, iron, zinc, cobalt, overall minerals, calcium, phosphorus, magnesium, sodium, potassium) grain corn was the methodology described in AOAC (1990). The analysis bacteria (total bacteria, coliforms at 35 °C to 45 °C coliforms, *Escherichia coli*, *Staphylococcus* sp., *Salmonella* sp., Sulphite reducing clostridia) was done using the general method described by the American Public Health Association American Public Health Association-APHA (SPECK, 1984). The analysis of toxigenic fungi (total fungi, *fusarium* sp., *aspergillus* sp., *aspergillus flavus*) was performed by DHINGRA & SINCLAIR (1996). The analysis of mycotoxins (aflatoxins, fumonisins, ocratoxins A, zearalenone, deoxynivalenol, nivalenol fusarenon X, T-2) in the corn samples was done using a high performance liquid chromatography (HPLC) method, following the detection method proposed by PERAICA et al. (2002). The variations in CO<sub>2</sub> levels were monitored daily, with the aid of a digital analyzer of CO<sub>2</sub>. The

CO<sub>2</sub> concentration readings were taken within the grains and the storage chambers (%) (VOGEL et al., 2002).

**Results and discussion**

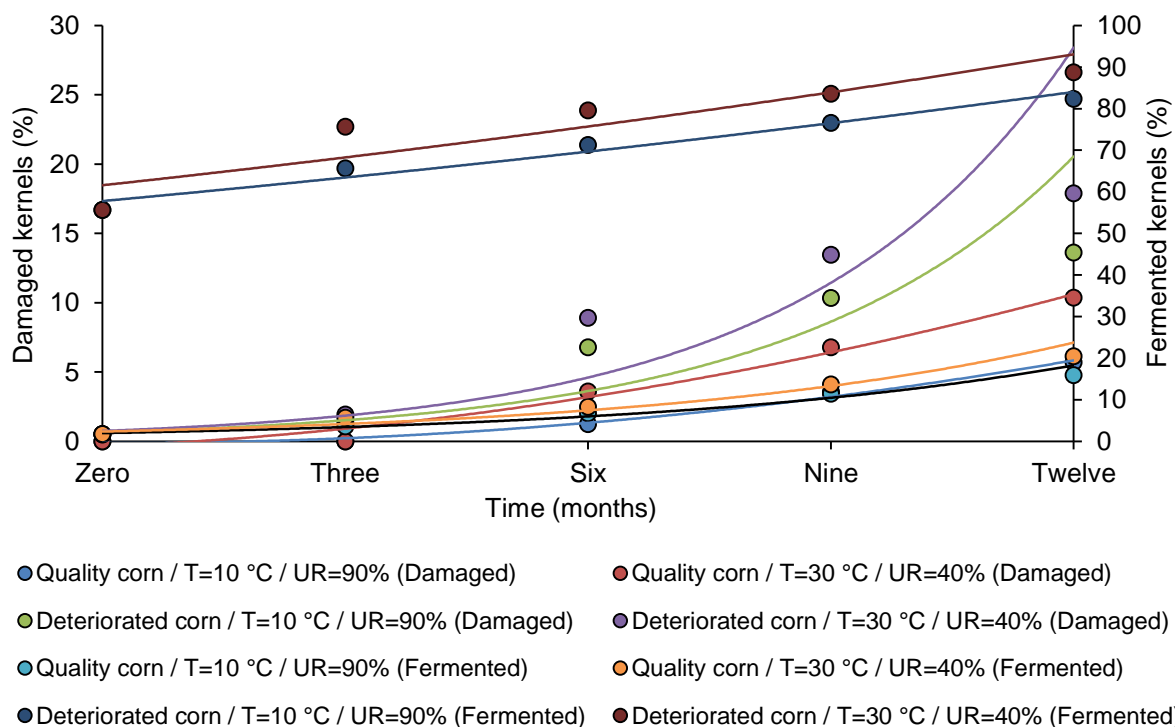
The physical classification is one of the important factors that characterize the quality of the corn trade. Table 1 is the physical quality of corn grain was stored over the twelve months. According LORINI et al. (2002), considered to be a product of the market requires that corn having a maximum water content of 14.5% (w.b.), content of impurities of 1% on the sieve of 3 mm or up to

3% in 5 mm sieve, maximum rate of 6% rot and avoid the presence of fungi and toxins. The corn grain for animal feed must be free from fungi, mycotoxins, toxic seeds and pesticide residues and should fit in types 1, 2 or 3, as the Ordinance of the Ministry of Agriculture Brazil (BRASIL, 2006), meeting the minimum attributes quality.

Figure 1 it was observed that the physical quality of corn reduced during the storage time (p<0.05), regardless of storage condition. The amounts of grain rot and fermentation conditions were higher than 30 °C and 40% relative humidity in storage.

**Table 1** - Physical classification of corn.

Physical classification	Corn quality	Deteriorated corn
Water content (w.b.)	12.43	17.61
Foreign matter (%)	1.30	1.60
Dross (%)	1.20	1.50
Fragments (%)	3.10	3.40
Broken (%)	7.30	8.40
Cracked (%)	2.60	6.80
Damaged (%)	4.80	9.20
Fermented (%)	15.10	29.70
Moldy (%)	0.00	10.50
Drilled (%)	13.60	15.10
Immature (%)	2.20	2.40
Live insects / seeds toxic	ND	ND
Class	yellow	yellow
Group	Hard	Hard



**Figure 1** - Damaged grains and fermented during storage (%).

The corn grain quality was less affected by storage conditions that damaged grains. In all treatments indices burnt grains and fermented become significant in lots, from the three months of storage ( $p < 0.05$ ). Throughout the storage time of corn was in all batches larger quantities of fermented beans than burned, but from six months storage the contents of damaged kernels of grain increased and remained constant fermented. The burning grains constitute a major problem in the warehouses of corn, due to falling grain quality, resulting in the depreciation of production. According to PINTO et al. (2007) and CORADI et al. (2011) starts the process of deterioration in the field and extends over the post-harvest processes may or may not undergo fermentation stages before the grains are burnt. According to these authors, the causal agents of rot grains most commonly found in corn are *Fusarium*, *Aspergillus*, *Penicillium*, *Cladosporium*, *Cephalosporium* and *Stenocarpella*.

Table 2 presents the models adjusted for the percentage of damaged grains and fermented

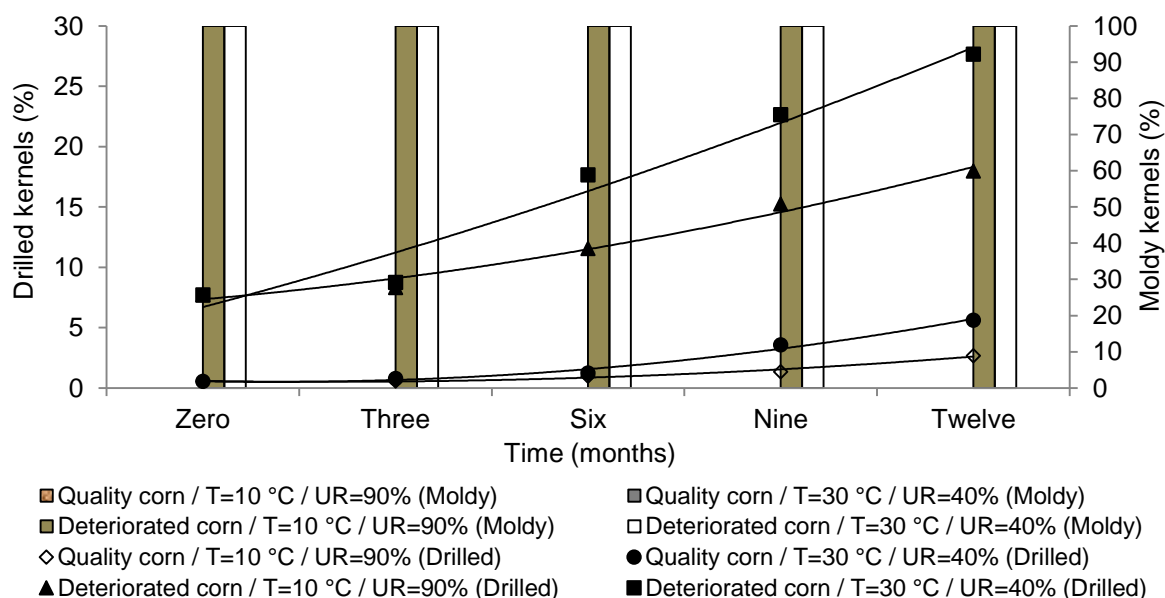
a function of time, the quality of corn and storage condition. The settings of exponential and polynomial models were adequate for treatments. It was noted that the models had a high significance level for the coefficient of determination ( $R^2$ ) and only the corn kernels fermented type deteriorated stored at 30 °C and relative humidity of 40% was not significant under the test F. Figure 2 was compared moldy grains and drilled over storage time for different conditions of temperature and relative humidity.

It was observed that there was an increase of grain drilled with increasing storage time, especially significantly ( $p < 0.05$ ) from the three months. For the grain drilled, it was found that the conditions of 30 °C and 40% more negatively influence the quality, whereas in the types of grain, deteriorated product was greatly affected. Moreover, the grains did not show quality problems with mildew in any of the storage conditions, over twelve months, whereas damaged grains got moldy and remained at all times, regardless of storage conditions.

**Table 2** - Equations fitted to the values of damaged grains and fermented for treatments of corns and deteriorated quality storage conditions of 30 °C and 40%, 10 °C and 90%.

Treatment	Equation	R <sup>2</sup> (%)
Deteriorated corn / T = 30 °C/ UR=40% (Fermented)	$y = 55.54 e^{0.1032x}$	80.7 <sup>NS</sup>
Deteriorated corn / T = 10 °C/ UR=90% (Fermented)	$y = 52.614 e^{0.0936x}$	96.3*
Deteriorated corn / T = 30 °C/ UR=40% (Damaged)	$y = 0.3006 e^{0.9097x}$	90.8*
Deteriorated corn / T = 10 °C/ UR=90% (Damaged)	$y = 0.2685 e^{0.8676x}$	91.7*
Quality corn / T = 30 °C/ UR=40% (Fermented)	$y = 0.3799x^2 - 0.7871x + 0.2783$	99.0**
Quality corn / T = 10 °C/ UR=90% (Fermented)	$y = 1.4925x - 2.3814$	90.8*
Quality corn / T = 30 °C/ UR=40% (Damaged)	$y = 0.4819x^2 - 0.1402x - 0.7317$	98.3**
Quality corn / T = 10 °C/ UR=90% (Damaged)	$y = 1.1259 e^{0.5575x}$	98.1**

\*\* Significant at 1%, the F test, \*Significant at 5%, the F test, <sup>NS</sup>Not significant.



**Figure 2** - Moldy grain and drilled over time storage (%).

In Table 3 there is the polynomial regression equations fitted to the experimental values of grain drilled and coefficients of determination ( $R^2$ ) with their significance levels. Analyzing the results of Table 3, it is seen that the polynomial equation representing satisfactorily experimental data.

Tables 4 and 5 are the physical-chemical evaluations of corn stored for the twelve months of storage. It was observed that there were no

significant differences in quality for most evaluations ( $p < 0.05$ ), with the exception of some minerals, such as iron, zinc, magnesium, sodium, potassium and calcium that varied over time, regardless of the type of stored corn. The increase of water content from the three months of storage had a negative influence on the quality of the stored product, mainly for grain corn of low quality.

**Table 3** - Equations adjusted to values of grain drilled and moldy grains for the treatment of corn quality deterioration and storage conditions of 30% and 40 °C, 10 °C and 90%.

Treatment	Equation	$R^2$ (%)
Deteriorated corn / T = 30 °C/ UR=40% (Drilled)	$y=0.2865x^2+3.607x+2.7546$	96.7*
Deteriorated corn / T = 10 °C/ UR=90% (Drilled)	$y=0.336x^2+0.7282x+6.2838$	98.4**
Quality corn / T = 10 °C/ UR=90% (Drilled)	$y=0.1838x^2-0.5981x + 0.9964$	97.9*
Quality corn / T = 30 °C/ UR=40% (Drilled)	$y=0.3939x^2-1.0696x+1.2254$	98.8**

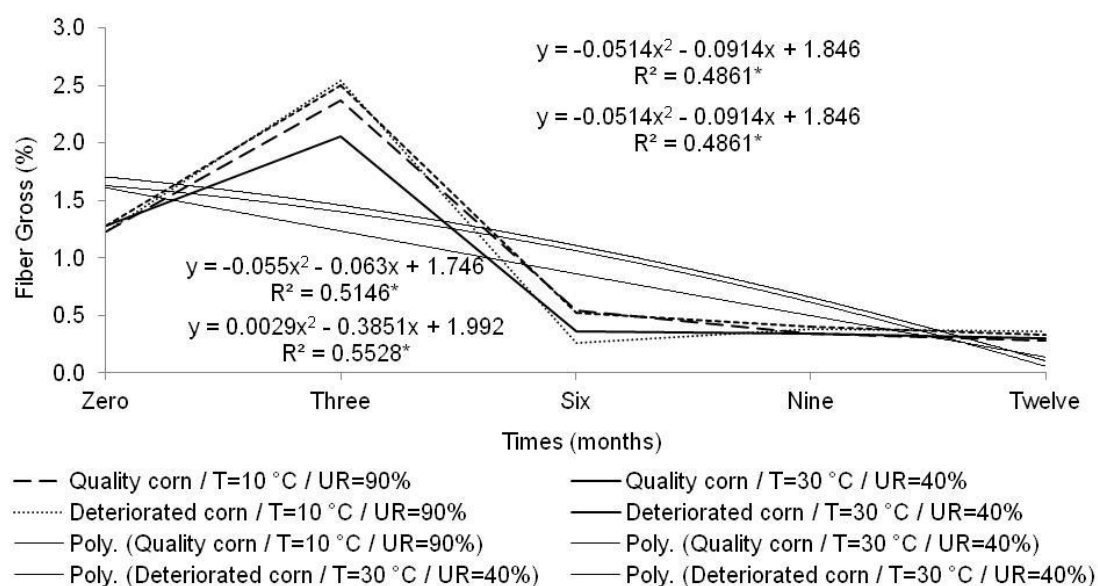
\*\* Significant at 1%, the F test, \*Significant at 5% by the F test, <sup>NS</sup> Not significant.

**Table 4** - Physical-chemical grain corn quality deteriorated and stored under different conditions of temperature and storage.

Analyses	Times (months)	Quality	Quality	Deteriorated	Deteriorated
		corn / T=10 °C / UR=90%	corn / T=30 °C / UR=40%	corn / T=10 °C / UR=90%	corn / T=30 °C / UR=40%
Water content (% w.b.)	Zero	12.43 aA	12.43 aA	17.61 bA	17.61 bB
	Three	13.56 bA	11.32 aA	17.16 cA	11.09 aA
	Six	13.93 bB	12.01 aB	17.04 cA	11.89 aA
	Nine	14.12 bB	12.56 aB	17.15 cA	12.13 aA
	Twelve	14.44 bB	12.98 aB	17.20 cA	12.38 aA
Dry matter (%)	Zero	87.57 bA	87.57 bA	82.39 aA	82.39 aA
	Three	86.44 bA	88.68 bA	82.84 aA	88.91 bB
	Six	86.07 bA	87.99 bA	82.96 aA	88.11 bB
	Nine	85.88 aA	87.44 bA	82.50 aA	87.87 bB
	Twelve	85.56 aA	87.02 bA	82.80 aA	87.62 bB
Extract not nitrogen (%)	Zero	74.27 bB	74.27 bB	69.15 aA	69.15 aA
	Three	73.17 aB	75.57 aB	72.97 aB	72.97 aA
	Six	72.88 aB	76.71 bB	71.59 aB	75.15 bB
	Nine	71.78 aA	75.68 bB	72.45 aB	74.98 bB
	Twelve	68.95 aA	70.14 aA	70.84 bA	72.54 bA
Copper (ppm)	Zero	20.00 aA	20.00 aA	20.00 aA	20.00 aA
	Three	20.00 aA	20.00 aA	20.00 aA	20.00 aA
	Six	20.00 aA	20.00 aA	20.00 aA	20.00 aA
	Nine	20.00 aA	20.00 aA	20.00 aA	20.00 aA
	Twelve	20.00 aA	20.00 aA	20.00 aA	20.00 aA
Manganese (ppm)	Zero	10.00 aA	10.00 aA	10.00 aA	10.00 aA
	Three	10.00 aA	10.00 aA	10.00 aA	10.00 aA
	Six	10.00 aA	10.00 aA	10.00 aA	10.00 aA
	Nine	10.00 aA	10.00 aA	10.00 aA	10.00 aA
	Twelve	10.00 aA	10.00 aA	10.00 aA	10.00 aA
Iron (ppm)	Zero	31.00 aA	31.00 aA	50.00 bB	50.00 bB
	Three	25.00 aA	26.00 aA	61.00 bB	35.00 aA
	Six	50.00 cB	25.00 aA	39.00 bA	39.00 bA
	Nine	32.00 aA	26.00 aA	38.00 bA	37.00 bA
	Twelve	28.00 aA	24.00 aA	30.00 bA	31.00 bA
Zinc (ppm)	Zero	16.00 aA	16.00 aA	17.00 aB	17.00 aB
	Three	11.00 aA	11.00 aA	11.00 aA	10.00 aA
	Six	25.00 bB	22.00 bB	17.00 aB	19.00 aB
	Nine	28.00 bB	24.00 bB	19.00 aB	21.00 aB
	Twelve	29.00 bB	26.00 bB	20.00 aB	23.00 aB
Cobalt (ppm)	Zero	20.00 aA	20.00 aA	20.00 aA	20.00 aA
	Three	20.00 aA	20.00 aA	20.00 aA	20.00 aA
	Six	20.00 aA	20.00 aA	20.00 aA	20.00 aA
	Nine	20.00 aA	20.00 aA	20.00 aA	20.00 aA
	Twelve	20.00 aA	20.00 aA	20.00 aA	20.00 aA

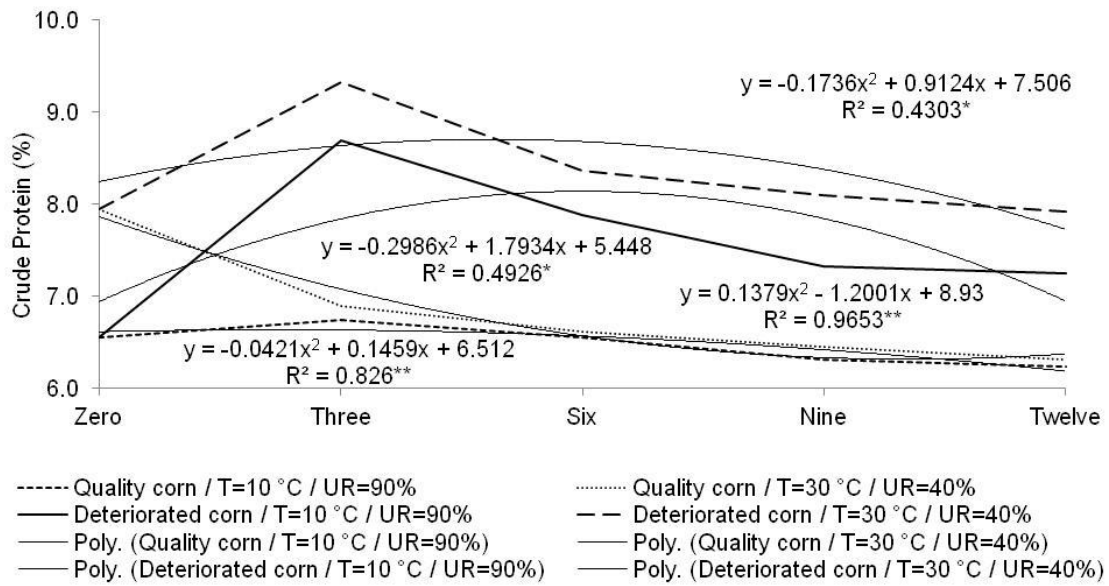
**Table 5** - Physical-chemical grain corn quality deteriorated and stored under different conditions of temperature and storage.

Analyses	Times (months)	Quality corn / T=10 °C / UR=90%	Quality corn / T=30 °C / UR=40%	Deteriorated corn / T=10 °C / UR=90%	Deteriorated corn / T=30 °C / UR=40%
Total Minerals (%)	Zero	1.05 bA	1.05 bA	0.91 aA	0.91 aA
	Three	0.96 bA	0.92 bA	0.82 aA	0.87 aA
	Six	1.08 bA	1.09 bA	0.90 aA	0.94 aA
	Nine	1.12 bB	1.16 bB	1.02 aB	1.06 aB
	Twelve	1.16 bB	1.20 bB	1.10 aB	1.13 aB
Calcio (%)	Zero	0.10 aA	0.10 aA	0.10 aA	0.10 aA
	Three	0.10 aA	0.10 aA	0.10 aA	0.10 aA
	Six	0.13 aB	0.12 aB	0.13 aB	0.13 aB
	Nine	0.14 aB	0.13 aB	0.14 aB	0.14 aB
	Twelve	0.12 aB	0.13 aB	0.13 aB	0.14 aB
Phosphorus (%)	Zero	0.29 bB	0.29 bB	0.23 aB	0.23 aB
	Three	0.27 bB	0.26 bB	0.22 aB	0.22 aB
	Six	0.26 bB	0.24 bB	0.19 aA	0.24 aB
	Nine	0.23 bA	0.22 bA	0.18 aA	0.19 aA
	Twelve	0.20 bA	0.19 bA	0.16 aA	0.16 aA
Magnesium (%)	Zero	0.09 aB	0.09 aB	0.09 aB	0.09 aB
	Three	0.09 bB	0.09 bB	0.08 aB	0.07 aB
	Six	0.07 bB	0.06 bA	0.05 aA	0.06 bA
	Nine	0.06 bA	0.05 aA	0.04 aA	0.04 aA
	Twelve	0.05 aA	0.05 aA	0.05 aA	0.04 aA
Sodium (%)	Zero	0.005 aA	0.005 aA	0.005 aA	0.005 aA
	Three	0.009 bB	0.005 aA	0.005 aA	0.005 aA
	Six	0.010 aB	0.010 aB	0.010 aB	0.010 aB
	Nine	0.020 aC	0.020 aC	0.020 aC	0.020 aC
	Twelve	0.020 aC	0.020 aC	0.020 aC	0.020 aC
Potassium (%)	Zero	0.31 aA	0.31 aA	0.26 bA	0.26 bA
	Three	0.27 aA	0.36 bB	0.28 aA	0.24 aA
	Six	0.38 bB	0.38 bB	0.25 aA	0.28 aA
	Nine	0.41 bB	0.40 bB	0.26 aA	0.30 aA
	Twelve	0.44 bB	0.41 bB	0.29 aA	0.34 aB



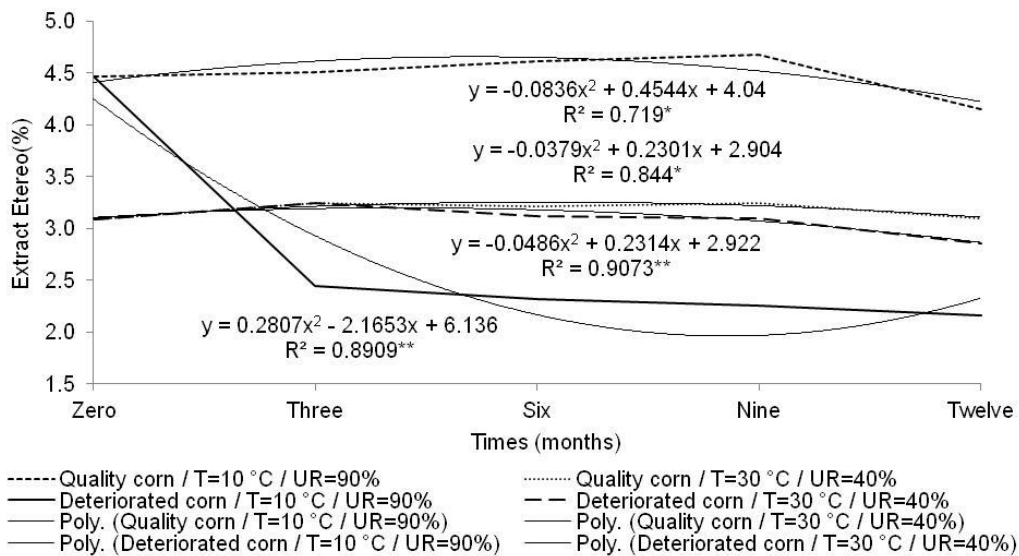
\*Significant at 5% by the t test.

**Figure 3** - Percent crude fiber in corn stored over time.



\*\* Significant at 1%, the F test. \*Significant at 5%, the t test.

Figure 4 - Crude protein in corn stored over time.



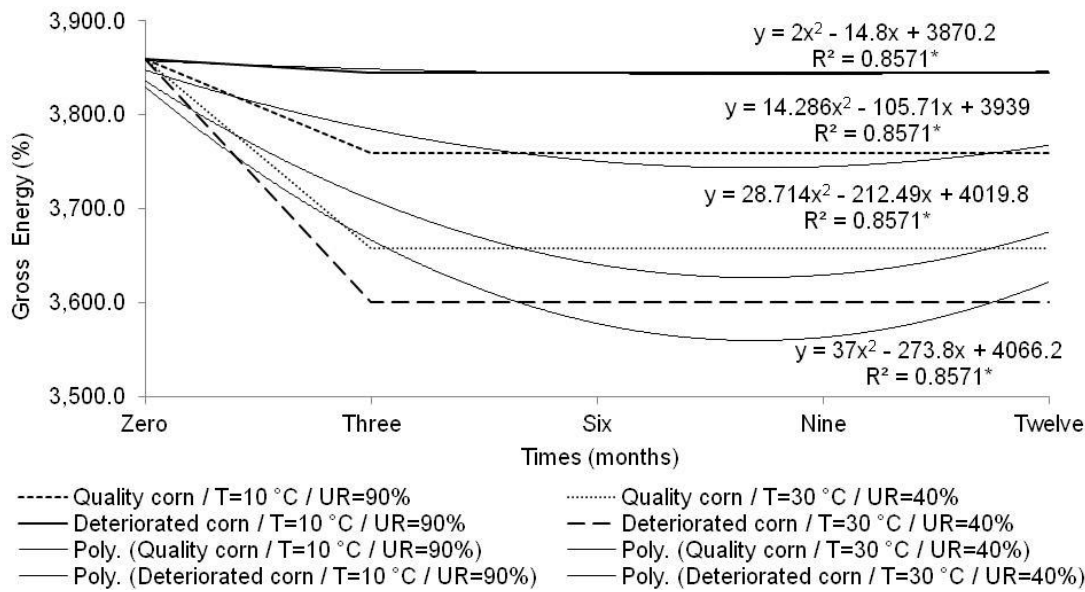
\*\* Significant at 1%, the t test. \*Significant at 5%, the t test.

Figure 5 - Percentage of extract etereo in corn stored over time.

The percent crude fiber varied significantly throughout the storage period ( $p < 0.05$ ), increased after three months, however, after six months lost quality and below the initial remaining virtually constant up to twelve months of storage. Among corns evaluated and storage conditions, no significant differences were observed.

The contents of crude protein and ether extract varied significantly in treatments and over storage time ( $p < 0.01$ ). According to the analysis results, the corn had deteriorated with lower levels of crude protein and ether extract than

corn quality, with a slight increase in rates in the three months of storage and reduction at twelve months. According to ANTUNES et al. (2011) insects first attack the endosperm of maize grains, though the insects consume more germ, causing a greater decrease in fat and protein may even increase for two reasons: first, for normal aspect ratio due to increased consumption of fat and less protein and, secondly, because the sample can be determined along with the grain protein, the protein of insects and, thus, can also increase the percentage.



\*Significant at 5%, the t test.

Figure 6 - Percentage of gross energy in corn stored over time.

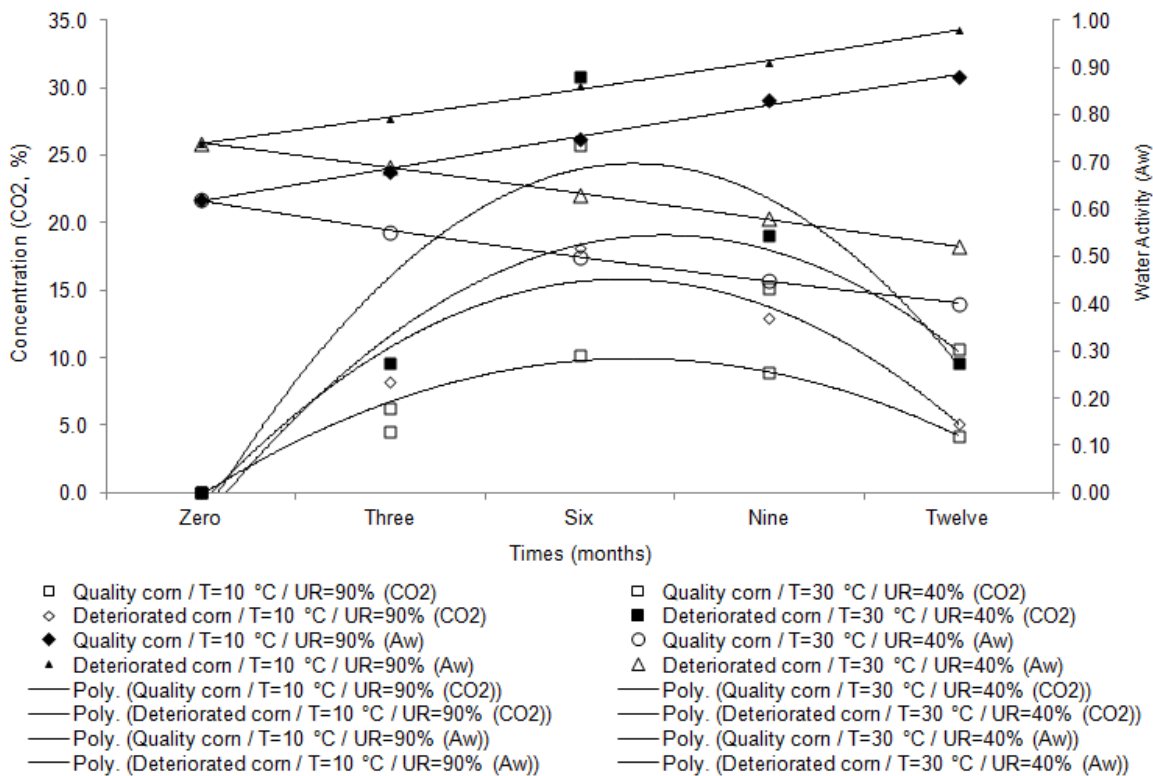


Figure 7 - CO<sub>2</sub> concentration (%) and water activity (Aw) in corn stored over time.

According to GODOI et al. (2008), when the corn is used for power can result in loss in animal performance and, ultimately, on the profitability of production. The speed and intensity of the loss depends on factors intrinsic quality of the grain, the operations of pre-storage, storage system used, and environmental conditions dur-

ing the storage period (ELIAS, 2000), agreeing with the results observed in this study. Thus, it can be said that the proteins of corn undergo reactions such as hydrolysis deamidation and complexation with other chemical components of the grains themselves, since the formation and remaining after harvest. According to some au-



thors, for the deamination of amino acids for the formation of organic acids and ammonium compounds. Already by decarboxylation, originated mainly amines, process characteristics of grains rot, producing strong odors and unpleasant. Complexation sugars occurs darkening of grains, with a consequent decrease in nitrogen content and increased protein content of non-protein nitrogen (RIBEIRO et al., 2005; ANTUNES et al., 2011). Figure 7 are the values of CO<sub>2</sub> and water activity of corn stored under different conditions of temperature and relative humidity.

According to Table 6, the values of Aw and CO<sub>2</sub> were significant at the 1% and 5% probability. The polynomial equations were

satisfactory adjustment of the experimental data. It has been observed in Figure 7, there was an increase in the breathing process of the corn grains after three months of storage, reducing its intensity at nine months of storage, with higher values for the damaged grains and the conditions 10 °C and 90% relative humidity. On the other hand the water activity of the corn grains tended to respiratory process of the product, ie the Aw was increased over time in stored grain storage conditions of 10 °C and 90% relative humidity, and higher Aw values for the corn grains deteriorated. While for the corn grains stored at 30 °C and 40% relative humidity Aw reduced over time, being lower Aw to corn quality.

**Table 6** - Equations adjusted to the concentrations of CO<sub>2</sub> and activity for the treatment of corns deteriorated in quality and storage conditions of 30 °C and 40%, 10 °C and 90%.

Treatment	Equation	R <sup>2</sup> (%)
Deteriorated corn / T = 30 °C/ UR=40% (CO <sub>2</sub> )	$y=-5.0664x^2+33.282x-30.294$	81.6*
Deteriorated corn / T = 10 °C/ UR=90% (Aw)	$y = 0.0014x^2+0.0514x+0.686$	99.8**
Deteriorated corn / T = 30 °C/ UR=40% (Aw)	$y=-0.0007x^2-0.0507x+0.792$	99.9**
Deteriorated corn / T = 10 °C/ UR=90% (CO <sub>2</sub> )	$y=-3.3757x^2+21.744x-19.218$	92.8*
Quality corn / T = 30 °C/ UR=40% (CO <sub>2</sub> )	$y=-3.5664x^2+24.582x-23.294$	70.2*
Quality corn / T = 10 °C/ UR=90% (Aw)	$y=-0.0007x^2+0.0713x+0.546$	99.6**
Quality corn / T = 30 °C/ UR=40% (Aw)	$y=0.0029x^2 -0.0711x+0.686$	99.8**
Quality corn / T = 10 °C/ UR=90% (CO <sub>2</sub> )	$y=-1.9386x^2+12.733x-10.982$	99.3**

\*\* Significant at 1%, the t test. \*Significant at 5%, the t test.

**Table 7** - Evaluation of fungus (CFU g<sup>-1</sup>) for different types of corn, conditions and time of storage.

Evaluation	Times (months)	Quality corn / T=10 °C / UR=90%	Quality corn / T=30 °C / UR=40%	Deteriorated corn / T=10 °C / UR=90%	Deteriorated corn / T=30 °C / UR=40%
Total fungi	Zero	1.3x10 <sup>1</sup>	3.3x10 <sup>3</sup>	1.3x10 <sup>1</sup>	3.3x10 <sup>3</sup>
	Three	1.3x10 <sup>5</sup>	1.5x10 <sup>4</sup>	6.2x10 <sup>5</sup>	5.7x10 <sup>5</sup>
	Six	2.7x10 <sup>6</sup>	1.9x10 <sup>6</sup>	8.02x10 <sup>7</sup>	8.02x10 <sup>6</sup>
	Nine	1.3x10 <sup>7</sup>	2.5x10 <sup>7</sup>	7.2x10 <sup>8</sup>	7.1x10 <sup>7</sup>
	Twelve	6.9x10 <sup>7</sup>	5.7x10 <sup>7</sup>	9.8x10 <sup>8</sup>	8.5x10 <sup>7</sup>
<i>Fusarium</i> sp.	Zero	0.4x10 <sup>1</sup>	2.5x10 <sup>3</sup>	0.4x10 <sup>1</sup>	2.5x10 <sup>3</sup>
	Three	1.2x10 <sup>3</sup>	5.3x10 <sup>2</sup>	5.5x10 <sup>3</sup>	5.1x10 <sup>3</sup>
	Six	2.4x10 <sup>4</sup>	7.1x10 <sup>4</sup>	1.2x10 <sup>5</sup>	0.6x10 <sup>4</sup>
	Nine	3.2x10 <sup>5</sup>	6.3x10 <sup>5</sup>	3.4x10 <sup>6</sup>	1.7x10 <sup>5</sup>
	Twelve	7.7x10 <sup>5</sup>	6.9x10 <sup>5</sup>	3.4x10 <sup>7</sup>	1.7x10 <sup>6</sup>
<i>Aspergillus</i> sp.	Zero	0.2x10 <sup>1</sup>	2.2x10 <sup>2</sup>	0.2x10 <sup>1</sup>	2.2x10 <sup>2</sup>
	Three	4.2x10 <sup>4</sup>	6.6x10 <sup>3</sup>	5.6x10 <sup>5</sup>	7.4x10 <sup>6</sup>
	Six	6.1x10 <sup>5</sup>	9.5x10 <sup>5</sup>	5.9x10 <sup>6</sup>	8.5x10 <sup>5</sup>
	Nine	5.2x10 <sup>6</sup>	8.4x10 <sup>6</sup>	6.7x10 <sup>7</sup>	6.3x10 <sup>3</sup>
	Twelve	8.0x10 <sup>7</sup>	7.4x10 <sup>7</sup>	7.6x10 <sup>8</sup>	7.9x10 <sup>7</sup>
<i>Aspergillus flavus</i>	Zero	0.1x10 <sup>1</sup>	1.5x10 <sup>2</sup>	0.1x10 <sup>1</sup>	1.5x10 <sup>2</sup>
	Three	2.0x10 <sup>3</sup>	5.7x10 <sup>3</sup>	2.2x10 <sup>4</sup>	4.7x10 <sup>4</sup>
	Six	1.8x10 <sup>4</sup>	3.8x10 <sup>4</sup>	1.2x10 <sup>6</sup>	4.8x10 <sup>5</sup>
	Nine	2.6x10 <sup>5</sup>	4.7x10 <sup>5</sup>	4.2x10 <sup>7</sup>	5.6x10 <sup>6</sup>
	Twelve	8.9x10 <sup>5</sup>	7.3x10 <sup>5</sup>	5.7x10 <sup>8</sup>	6.6x10 <sup>7</sup>

The respiratory rate has been used by several authors to estimate the loss of grain dry matter during storage (ALVES et al., 2006; SANTOS et al., 2012). In these studies, the respiratory rate was expressed as the ratio between the mass of carbon dioxide (CO<sub>2</sub>) produced dry matter and grain. In the proposed models, the production of CO<sub>2</sub> was converted into dry matter loss of grain considering the process of aerobic respiration with the complete oxidation of carbohydrates to CO<sub>2</sub>, water (H<sub>2</sub>O) and heat. In this context, production of 14.7g of CO<sub>2</sub> per kg dry matter of grains equals 1% loss of dry matter (SAUL & STEELE, 1966). According DILLAHUNTY et al. (2000), the factors of moisture content of grain, index of mechanical damage, temperature and grain storage environment and the composition of the atmosphere, especially the availability of O<sub>2</sub>, influence the respiratory activity of the grains and consequent loss of dry matter, results associated with producing fungi and insect proliferation.

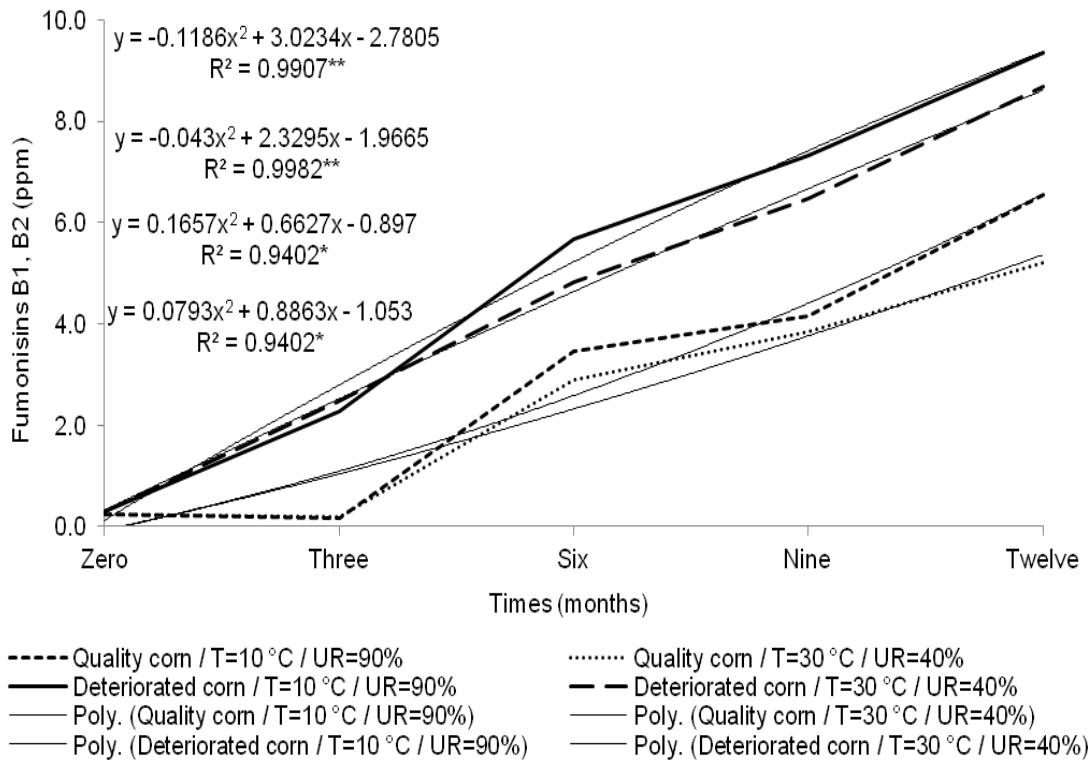
Thus, the results in Table 7, it was found

that the growth of colonies of fungi (*Aspergillus flavus*) increased during storage time of corn from 0.1x10<sup>1</sup> to 5.7x10<sup>8</sup>. Consequently, it was found that corn was more susceptible to deteriorated fungal infection (9.8x10<sup>8</sup>), whereas the storage conditions of low temperature and high relative humidity accelerated the development of *Aspergillus* sp. (0.2x10<sup>1</sup> to 7.6x10<sup>8</sup>) and *Fusarium* sp. (0.4x10<sup>1</sup> to 3.4x10<sup>7</sup>), agreeing with the results of BOHRA & PUROHIT (2003).

It was found in Table 8, which over storage time there was a increased number of bacterial colonies from 2.5x10<sup>3</sup> to 4.8x10<sup>7</sup>. Among the bacteria, the most observed were *Coliformes* at 35 °C (<10) and *Staphylococcus* sp. (<100). Were not observed contamination by *E. coli* and low contamination by *Salmonella* sp. A relatively small number of studies have examined the presence of *Salmonella* sp. and *E. coli* during the manufacturing processes of feeds, especially in light of flour and animal by-products mixed into the final feed formulation.

**Table 8** - Evaluation of bacteria (CFU.g<sup>-1</sup>) for different types of corn, the conditions and time of storage.

Evaluations	Times (months)	Quality corn / T=10 °C / UR=90%	Quality corn / T=30 °C / UR=40%	Deteriorated corn / T=10 °C / UR=90%	Deteriorated corn / T=30 °C / UR=40%
Total bacteria	Zero	2.5x10 <sup>3</sup>	9.1x10 <sup>4</sup>	2.5x10 <sup>3</sup>	9.1x10 <sup>4</sup>
	Three	8.1x10 <sup>4</sup>	9.4x10 <sup>2</sup>	1.9x10 <sup>7</sup>	4.1x10 <sup>5</sup>
	Six	2.7x10 <sup>2</sup>	4.8x10 <sup>4</sup>	2.5x10 <sup>3</sup>	2.4x10 <sup>5</sup>
	Nine	4.7x10 <sup>4</sup>	3.5x10 <sup>6</sup>	4.1x10 <sup>5</sup>	4.2x10 <sup>6</sup>
	Twelve	5.8x10 <sup>5</sup>	4.8x10 <sup>7</sup>	5.3x10 <sup>6</sup>	7.5x10 <sup>6</sup>
Coliformes a 35 °C	Zero	<10	<10	<10	<10
	Three	<10	<10	5.3x10 <sup>3</sup>	<10
	Six	<10	<10	<10	<10
	Nine	<10	<10	<10	<10
	Twelve	<10	<10	<10	<10
Coliformes a 45 °C	Zero	<10	<10	<10	<10
	Three	<10	<10	20	<10
	Six	<10	<10	20	<10
	Nine	<10	<10	20	<10
	Twelve	<10	<10	20	<10
<i>Escherichia coli</i>	Zero	<10	<10	<10	<10
	Three	<10	<10	20	<10
	Six	<10	<10	20	<10
	Nine	<10	<10	20	<10
	Twelve	<10	<10	20	<10
<i>Staphylococcus</i> sp.	Zero	<100	<100	<100	<100
	Three	<100	<100	<100	<100
	Six	<100	<100	<100	1.8x10 <sup>4</sup>
	Nine	<100	<100	<100	2.9x10 <sup>6</sup>
	Twelve	<100	<100	<100	4.7x10 <sup>6</sup>
<i>Salmonella</i> sp.	Zero	ausent 25g	ausent 25g	ausent 25g	ausent 25g
	Three	ausent 25g	ausent 25g	ausent 25g	ausent 25g
	Six	ausent 25g	ausent 25g	ausent 25g	ausent 25g
	Nine	ausent 25g	ausent 25g	present 25g	ausent 25g
	Twelve	present 25g	ausent 25g	present 25g	ausent 25g
<i>Clostridium sulfito redutor</i>	Zero	<10	<10	<10	<10
	Three	<10	<10	<10	<10
	Six	<10	<10	<10	<10
	Nine	<10	<10	<10	<10
	Twelve	<10	<10	<10	<10



\*\* Significant at 1%, the F test. \*Significant at 5%, the F test.

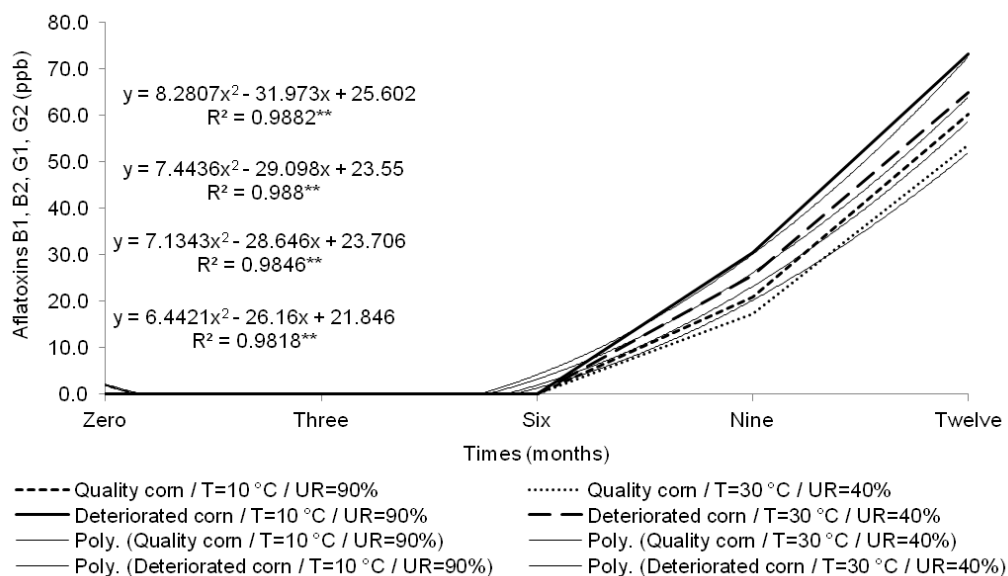
**Figure 8** - Quantitation of fumonisins (ppm) for different types of corn, the conditions and time of storage.

According to Figure 8, it was found that the results of aflatoxins and fumonisins in corn kernels were stored for significant treatments performed 1 and 5%. The coefficient of determination  $R^2$  is greater than 94% and most treatments were above 98%, ie satisfactory. In the initial conditions of storage, there was the presence of fumonisins in corn deteriorated (0.3053 ppm) and quality (0.2428 ppm).

Over time the storage was increased aflatoxins (73.26 ppb) (Figure 9). Aflatoxins and fumonisins were higher storage conditions with low temperature and high relative humidity (10 °C and 90%). This probably was due to increased infection of toxigenic fungi species *Aspergillus* sp. and *Fusarium* sp. The fungal contamination in vegetable substrates (corn) is favored with increasing water activity of the product (Figure 7), ie, when the external conditions of temperature and relative humidity imply changes in grains, mainly increasing the moisture balance of the product. However, according RAMAKRISHNA et al. (1996), whole grain is not necessarily contaminated by mycotoxins, since the production and concentration of these substances are determined by the combined effects of fungal species present, the temperature and the humidity of the grain. Thus, one may have fungus growing

in grains without any production of toxins (BROGGI et al., 2002; BRERA et al., 2004). In the United Nations Food and Agriculture Organization (FAO) estimates that 25% of the world grain contaminated with mycotoxins (REDAÇÃO, 2002), which is a value considerably higher. According PERAICA et al. (2002), the raw materials and derivatives are generally more contaminated with a mycotoxin, because some fungi are capable of producing different kinds of chemicals. However, this information was not confirmed in this study (Table 9).

The presence of mycotoxins in corn in the feed and therefore can affect animals and bring them to a number of complications in chickens, for example, the most common effects are increased and yellowing of the liver, kidney swelling, oral lesions, increased susceptibility to skin lesions, reduced pigmentation, increased susceptibility to infectious and parasitic diseases, impaired posture and change in the quality of their eggs, decreased weight gain and lower feed (FUNDAÇÃO ABC, 2010). These changes cause large losses, but often these are not accounted for, mainly due to lack of information of the creators. In Table 9 there was a measured result of some mycotoxin in corn, in which the presence was not identified.



\*\* Significant at 1%, the F test. \*Significant at 5%, the F test.

**Figure 9** - Quantification of aflatoxins (ppb) for different types of corn, the conditions and time of storage.

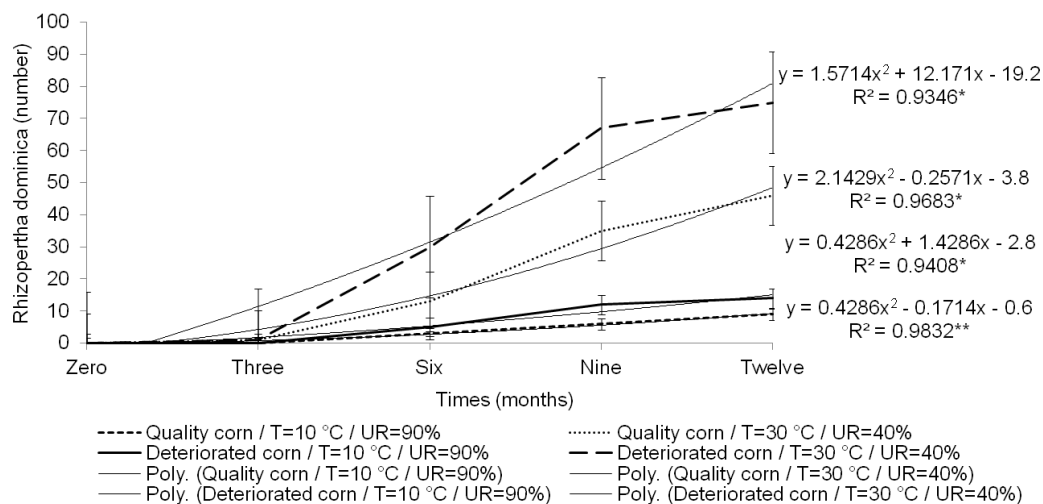
**Table 9** - Quantification of mycotoxins for different types of corn, the conditions and time of storage.

Evaluations	Times (months)	Quality corn / T=10 °C / UR=90%	Qualitycorn / T=30 °C / UR=40%	Deteriorated corn / T=10 °C / UR=90%	Deteriorated corn/ T=30 °C / UR=40%
Ocratoxins A	Zero	ND	ND	ND	ND
	Three	ND	ND	ND	ND
	Six	ND	ND	ND	ND
	Nine	ND	ND	ND	ND
	Twelve	ND	ND	ND	ND
Zearalenone	Zero	ND	ND	ND	ND
	Three	ND	ND	ND	ND
	Six	ND	ND	ND	ND
	Nine	ND	ND	ND	ND
	Twelve	ND	ND	ND	ND
Deoxynivalenol	Zero	ND	ND	ND	ND
	Three	ND	ND	ND	ND
	Six	ND	ND	ND	ND
	Nine	ND	ND	ND	ND
	Twelve	ND	ND	ND	ND
Nivalenol	Zero	ND	ND	ND	ND
	Three	ND	ND	ND	ND
	Six	ND	ND	ND	ND
	Nine	ND	ND	ND	ND
	Twelve	ND	ND	ND	ND
Fusarenon X	Zero	ND	ND	ND	ND
	Three	ND	ND	ND	ND
	Six	ND	ND	ND	ND
	Nine	ND	ND	ND	ND
	Twelve	ND	ND	ND	ND
T-2	Zero	ND	ND	ND	ND
	Three	ND	ND	ND	ND
	Six	ND	ND	ND	ND
	Nine	ND	ND	ND	ND
	Twelve	ND	ND	ND	ND

Figures 10, 11 and 12 shows the results for the identification and enumeration of insect pests of corn storage. The results obtained were high levels of insect infestation, significant increase ( $P < 0.01$ ), during the storage time, and the treatments with corn and deteriorated conditions of 30 °C and 40% relative humidity showed the worst results agreeing with SCHÖLLER et al. (1997), where losses of stored products identified as 30% in some cases, 10% of which are directly caused by the attack of pests (*Rhizopertha dominica*) during storage. The attack *Rhizopertha dominica* to stored grains, besides the quantitative losses arising from direct feeding of insects, expressive qualitative losses are driven, such as decreased nutritional value of grain and seed physiological quality which determines thus reducing the value market or even the condemnation of lots of seeds and / or grains, in agreement with the values obtained in Tables 3, 4, 5 and 6. According to SILVA et al. (1995) for processing or consumption, the value of the grain

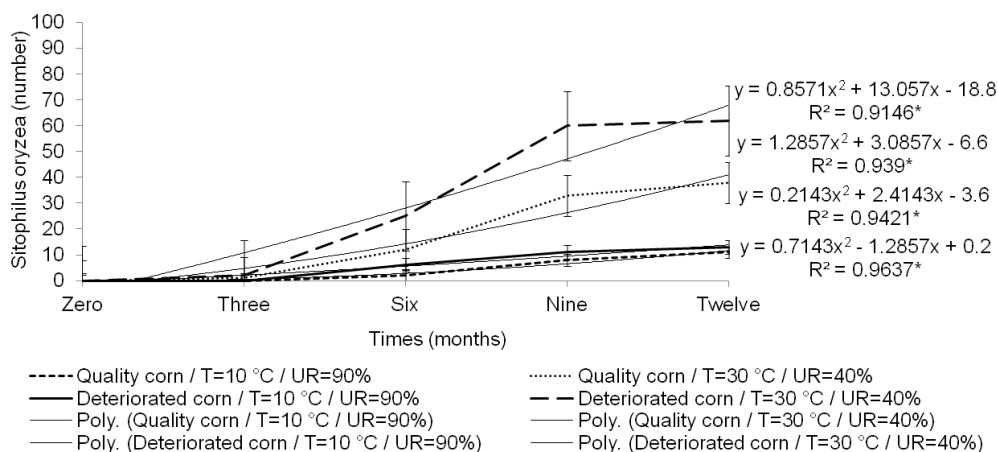
is directly related to the level of contamination by insects. The same author states that quantitative losses refer to decrease volume and weight, showing no degradation properly nutritional product.

*Sitophilus zeamais* (Motschulsky) (Coleoptera: Curculionidae), popularly known as weevil, is considered one of the most important pests in storage industry tropics (SILVEIRA et al., 2006). Its main features are: high biotic potential, ability to attack grain in the field and in storage units and survive at great depths in the grain mass. This type of insect was found at high levels in grains of corn stored in treatments (Figure 11) and the results are similar CANEPPELE et al. (2003) who studied the correlation between the level of infestation of *S. zeamais* and storage quality of corn, found increased weight loss of these grains, as were increased over time and the number of insects on contact with the corn kernels.



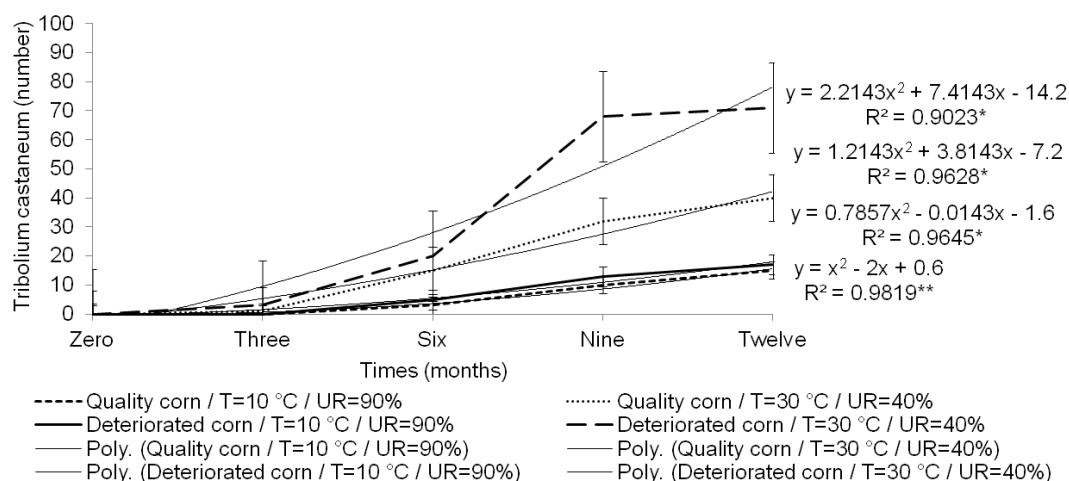
\*\* Significant at 1%, the F test. \*Significant at 5%, the F test.

Figure 10 - Identification and counting *Rhizopertha dominica* in stored corn.



\*\* Significant at 1%, the F test. \*Significant at 5%, the F test.

Figure 11 - Identification and enumeration of *Sitophilus oryzae* in stored corn.



\*\* Significant at 1%, the F test. \*Significant at 5%, the F test.

**Figure 12** - Identification and counting of *Tribolium castaneum* in stored corn.

In the same proportions of other insects were observed with the infestation on *Tribolium castaneum* corn stored infestation presented significant increase over time, whereas conditions 30 °C and 40% relative humidity were more favorable for the development *Tribolium castaneum* and the corn was damaged in the most affected. According FARONI et al. (2009) *T. castaneum* has a preference for flour and bran, but can attack wide variety of cereal grains and animal feed, especially when these products are with high moisture content and / or when the presence of decay fungi. This insect attacks all tops milled grains such as bran, broken grains, defective or already attacked by other pests. Their presence is a sign that the grains are infested by primary pests.

### Conclusions

The storage of corn grains in regions of warm, dry air; reduce the risk of incidence of fungi, regardless of initial quality of the stored product. On the other hand, the incidence of insect was greater for the same conditions. Regardless of storage conditions, bacterial growth was directly related to the quality of the initial product and the storage time.

Over time increased the risk of aflatoxins and fumonisins, reaching levels above acceptable limits (20 ppb and 5 ppm, respectively) for the quality of stored corn. The type of corn influenced stored in quality, although the proposed storage conditions were not appropriate to maintain the physical quality of corn up time of three months' storage.

To maintain the nutritional quality of corn in the conditions of 30 °C and 40% RH air for animal feed, it is recommended to store it in later than six months, while in regions with average temperatures of 10 °C and 90% RH are not recommended storing corn for a period exceeding three months.

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