IMPACT OF SEED COTTON COMPRESSION ON COTTONSEED QUALITY

A Thesis

by

NUR AZUAN BIN HUSIN

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Chair of Committee,	J. Alex Thomasson
Committee Members,	Yufeng Ge
	Gaylon D. Morgan
Head of Department,	Stephen W. Searcy

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ABSTRACT

Cotton is harvested as seed cotton, which includes fiber attached to seeds mixed with extraneous matter like leaves and stems. A module builder is used to compress the seed cotton into large rectangular modules that allow in-field storage until being transported to a ginning facility, where the fiber and seed are separated and cleaned. On-board module-building systems are now being used on harvesters and offer virtually continuous harvesting, without the need to transfer seed cotton to a separate module builder. With their smaller size and shape, these packages could possibly be transported more economically by loading multiple packages on a truck. However, their density is similar to traditional modules, so any transport advantage is minimal. Transportation costs could be reduced significantly by creating higher-density cotton packages at harvest. Compression of seed cotton to levels observed with module-building systems has not proven to damage cottonseed, but some research has shown that higher compression levels could cause damage. Moisture content and storage duration may also influence the effect of compression on the seed. The objective of this research was to quantify the effects of these factors, individually and in combination, on cottonseed across two cotton varieties. Humidity chambers were used to achieve desired experimental seed cotton moisture levels. A miniature bale press (16.6 x 8.3 x 6.2 in.) was used to compress bales of seed cotton to different densities. Germination and seed crackage were quantified to determine the impact on the cottonseed. Data analyses indicated that compression density, moisture content, storage time and position of the

sample in the bale were all significantly related to seed damage; however, cotton variety was not significantly related. Moisture was most strongly related to reduction in germination, while compression density was most strongly related to increasing crackage. Compression above 24 lbs/ft^3 was clearly associated with higher percentages of cracked seed.

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INTRODUCTION AND LITERATURE REVIEW

Cotton is one of the most important crops in the United States. The U.S. is ranked third in cotton production in the world, and Texas is its leading state (USDA, 2010). In addition to cotton fiber use in the textile industry, cotton's seeds are used for vegetable oil and animal feed. Seed quality is an important factor in cotton production when the seeds will be either replanted or used for oil extraction. Healthy, vigorous, and high-viability seeds are important for establishing good stands of cotton plants and subsequently for high yields and high fiber quality (Delouche, 1981). Cottonseed quality can be measured by emergence and survival of seedlings in various planting conditions. Injured or deteriorated cottonseed reduces the ability of seeds to overcome stresses (Comer, 1968). Water inside the seed the can seep through cuts and cracks in the seed coat, reducing germination potential. Injured seeds also attract more insects, adding to the potential for further injury (Stewart et al., 2009). Wilkes (1978) defined seed quality in terms of uniformity among the seeds, particularly with regard to germination percentage.

A concern about preserving seed quality relates to how seed cotton is stored prior to ginning. Various methods have been used to transport and store the harvested cotton: wagons, trailers, baskets, storage houses, turnrow storage, and the ricking system. In 1971, Texas A&M University and Cotton Incorporated developed the "module system" for handling and storing seed cotton that involved mechanical compression. Moduling produced higher storage densities (224 kg m^{-3}, 14 lb ft^{-3}) and provided a relatively weather-resistant package of seed cotton that could be stored in the field for fairly long times with little loss in fiber quality (Force, 2002). The move to moduling disengaged harvesting from ginning, so harvesting could proceed independent of the speed of ginning. The advantages of the module system have been widely accepted throughout the U.S., and by 2000 nearly all cotton farms were using the module system (Hughs et al., 2008).

The level of cotton production has remained fairly steady over the past 40 years (Hamann, 2011), but the number of gin facilities has decreased drastically. Thus, module trucks now have to travel farther, increasing fuel, maintenance, and labor costs. This issue has been a growing concern to farmers and gin managers and encouraged them to make modules as heavy as possible. Simpson et al. (2004) discussed the problem of overweight and oversize modules that resulted in fines and noted that special permits for oversize loads are costly. Recently, new cotton pickers have come to market with on-board packaging systems, offering the potential for continuous harvesting, and eliminating the need for separate module builders, boll buggies, and associated tractors and operators needed to operate the machinery. With their smaller size and shape, these

new packages may be able to be transported more economically by loading multiple packages on a truck, but their density is roughly the same as traditional modules, so any transport advantage would be minimal. It is clear that transportation costs could be reduced significantly by creating higher-density cotton packages at harvest. Compression of seed cotton to levels seen in traditional modules and on-board harvester packages has not proven to damage seed quality, but some research has shown that higher compression could damage the seeds (Lalor et al., 1995).

Moisture Content, Temperature, Storage

Seed cotton, a mixture of fiber and seed, absorbs moisture from or releases it to its environment depending on humidity and temperature conditions. The importance of moisture in seed cotton storage and handling has been noted by numerous past researchers (Anthony, 2004; Jaime et al., 2013; Parker and Wooten, 1964; Valco et al., 2004). Fiber quality tends to decrease during storage, and the effect is exacerbated by high moisture content (Wooten and Montgomery, 1956). Seed quality can also decrease during storage. Changes in seed cotton moisture content, with a storage density of 320 kg m^{-3} (12 lb ft^{-3}), have been studied (Sorenson and Wilkes, 1973) and an inverse relationship existed between moisture content and safe storage time (Figure 1). Abernathy and Williams (1961) studied the effects of baling seed cotton with a hay baler and found that bales could be stored up to 2 months, provided that the moisture content was less than 10%.

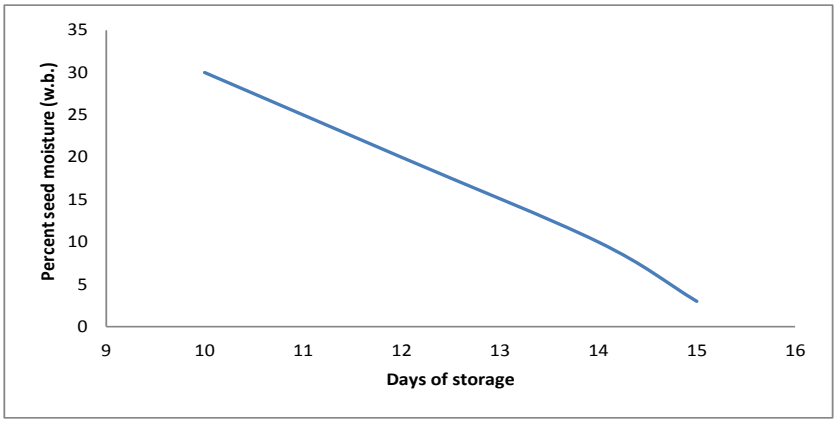

Figure 1. Relationship between moisture content and safe storage time (Sorenson and Wilkes, 1973).

Two indicators of decreasing seed quality in relation to high moisture content are reduced germination and increased free fatty acid content (Wilkes, 1974). Causes of high moisture content in seed cotton include high-moisture foreign matter, wet and humid weather, and wet storage conditions (Griffin Jr., 1974; Parker and Wooten, 1964; Shaw and Franks, 1962; Sorenson and Wilkes, 1959; Wilkes, 1978; Wang et al., 2010). A combination of warm ambient temperatures and long storage periods has been shown to reduce seed germination (Lalor et al., 1995).

Cottonseed can be used for planting, compressed for vegetable oil, and used as an animal feed (Jaime et al., 2013; Lichtenstein, 1990). Less oil and fewer nutrients can be extracted from low-quality seed. Fatty acid content in seed tends to increase at higher moisture levels in the cottonseed (Jaime et al., 2013). The acids are toxic at high levels, so cottonseed at high acid levels is not safe as a cattle feed or cooking oil. High moisture levels also enable microbial and fungal activity to increase storage temperature,

potentially resulting in membrane damage and enzyme deactivation (Hardin, 2004; Parker and Wooten, 1964). The effects of high temperature on cottonseed can result in poor cool germination test results (Jividen, 1986).

Wilkes (1978) determined that seed cotton compressed to densities ranging from 112 to 320 kg m^{-3} (7 to 20 lb ft^{-3}) with seed moisture levels less than 10% could be stored up to 30 days with no decrease in seed quality. In addition, it was found that cotton lint can withstand higher levels of moisture during storage compared to the seed. It was also reported that seed cotton densities from 80 to 112 kg m^{-3} (5 to 7 lb ft^{-3}) showed less effect on seed quality when moisture ranged from 10 to 12%. Seed cotton compression to a density of 400 kg m^{-3} (25 lb ft^{-3}) has been reported to physically damage the cottonseed (Lalor et al., 1995). Brashears et al. (1970), in their study on the pressure-density relationship with cottonseed quality, showed that seed cotton can be compressed up to 320 kg m^{-3} (20 lb ft^{-3}) without significantly damaging the seed. Previously, there has been little research on the effect of compression density associated with moisture and length of storage.

Mechanical Injury in Harvesting and Ginning

Physical damage is considered one of the most serious problems of seed production. Mechanical injury undergone by cottonseed can start at harvest and increase through ginning and delinting. Small gaps and low tolerances between the spindle and doffer in a cotton picker may cause damage to the seed coat. High picking speeds together with high fan speeds also increase the percentage of cracked seed (Colwick,

1972). The separation and cleaning processes on a stripper harvester can also induce damage to cottonseed (Douglas et al., 1967; Kılıçkan and Güner, 2006).

Excessively high ginning rates can also cause seed damage (Anthony and Mayfield, 1995). Tight seed-roll operation between the rotating gin saws and stationary ribs can inflict significant damage to the seed. Watson and Helmer (1964) found that increases in ginning rate caused increases in seed damage and reduced germination. Pneumatic handling systems in harvesters and the gin can cause damage to cottonseed as seeds sustain impact damage, striking walls at turns in pipes because of high air velocities in these systems.

With modern mechanical planting and cultural practices, flowability of the seeds is important to enable lower seeding rates. Before acid delinting was introduced, mechanical delinting or reginning operations and flame delinting were used to remove the lint and fibers on fuzzy seed. In mechanical delinting, damage could occur from the fine and closely spaced saws (Gelmond, 1979), while in flame delinting the seed could be damaged by the heat. These methods did not improve seed flowability sufficiently for mechanical planting (Delouche, 1981), so acid delinting became the procedure of choice, because it completely removes linters from the seed. The problem with acid delinting is the damage that can be caused by direct contact between the seed and a very reactive chemical, especially if the seed coat already has cuts as a result of mechanical injury. Other factors that may affect cottonseed quality are insects, over drying, impacts against other foreign objects like stones and debris, and worn or damaged machines and equipment.

Schoorl and Holt (1983) discussed the damage sustained by several types of seed and grain due to compression, which typically caused cracking, mostly observed only in dry seed. This mechanical damage results in lower germination and poor seedling growth. According to Kılıçkan and Güner (2006), it is necessary to make a miniature bale press to accurately represent the effect of compression on seed in a cotton module. This is true because instruments used to monitor compressive factors in a model press are typically more accurate than those that would be used in a full-size press. Furthermore, determining the behavior of individual cottonseed would not provide representative data for modules where bulk pressures are exerted.

Test to Evaluate Cottonseed Quality

Different tests are used to assess the suitability of cottonseed for planting purposes. The tests assess quality features of seed such as deterioration, germination potential, vigorousness and viability. Certain tests measure the biochemistry in the seed, including the tetrazolium test and free-fatty acid test. Gravity separation and cutting tests are used to measure the physical properties of the seed such as density and seed embryo color, respectively.

Delouche (1981) discussed different types of cottonseed injuries and how they can be distinguished by close visual examination. Typically seed damaged during harvesting exhibits cracked or straight fractured edges, and fragments of the seed coats are often missing, exposing the embryo. Seed damage during saw ginning involves cuts and deep gashes in the seed coat with rolling of the cut edges. The visual mechanical

damage test is used to evaluate the physical damage of the seed (Bewley et al., 2006). Manual detection with magnification devices to examine cracks and cuts on seed has been used before in many studies including those of Brashears et al. (1970) and Jividen (1986). This test is used to evaluate seed quality especially where seeds are to be acid-delinted. A magnification device and good lighting are necessary to visually classify damage severity as shown in Table 1 (Colwick, 1972). Douglas et al. (1965) and Douglas et al. (1967) used 100 subsamples of acid delinted seeds in their studies on cottonseed damage by a mechanical harvester. The damage grade ranged from "0" for no visible damage up to "4" for broken seed. Undamaged seeds are not adversely affected by acid-delinting, but severely damaged seeds usually do not remain viable after delinting, and minimally damaged seeds may germinate but are generally of low quality (Delouche, 1981).

Table 1. Damage seed classifications (Colwick, 1972).

Type of damage	Descriptions
No damage	Seeds with completely intact seed coats
Pinhole damage	Seeds with only one or two small punctures (pinhole) in seed coats.
Minor damage	Seeds with seed coats cracked or cut, but not severely. Damage primarily to the chalazal end or on sides of the seed.
Major damage	Seeds with large cuts or ruptures in the seed coats. Part of the seed coats missing, cotyledons exposed, or radicle end of the seeds damaged.

The germination test provides the most acceptable index of seed quality and possibly the most important indication of seed quality (Copeland, 1995). The test directly measures the germination potential by evaluating seed viability and vigorousness under favorable germination conditions of temperature, moisture and light. The germination rate is a good indicator of how well the seed will perform in the field.

The two predominant germination tests are the standard (warm) test and the cool test. The warm test follows the Association of Official Seed Analysts (AOSA, 1983) protocol: 8 replications of 50 seeds each for 12 days; seeds are placed in germination paper within a chamber; two alternating temperature and light regimes are used to simulate the diurnal cycle; normal seedlings are removed at days 4 and 8; seedlings a minimum of 1.5 inches long from tip of radicle to point of cotyledon attachment are considered normal. The cool test (most widely used in the U.S.) involves 4 replications of 50 seeds each; seeds are placed in germination paper within a chamber in the absence of light for seven days at 18°C (64° F). The main purpose of the cool test is to determine if seed lots are suitable for planting below ideal conditions such as cool soils. Hopper et al., (1988) reported that under adverse field conditions, the standard germination test does not adequately predict seed germination. Hopper also reported that studies have shown the cool test to be a better predictor of seed performance under field conditions than the standard germination test.

Detection of Damaged Cottonseed with Machine Vision

Machine vision has been used to detect cracks and other damage in corn, soybeans, wheat, rice, and other seeds (Luo et al., 1999; Wan et al., 2002; Zhang et al., 2007). Several researchers from universities in China have used the machine vision technology to detect the damage on cottonseed. Tongzhen and Li (2010) separated the broken seed from good seed using the binary images of cottonseed, while Shaojun and Ku (2009) separated the undamaged seeds from broken and cracked seeds based on the differences in the pixels on the edge of the seeds. Tao et al. (2009) used morphology to detect surface damage on cottonseed by analyzing cottonseed curvatures of contour points and seed-symmetry-boundary profile, while Li et al. (2012) also used a circularity parameter as the characteristic identifier of crushed cottonseed. Jing-bin et al. (2011) improved the method and used it for cottonseed variety identification using a back-propagation neural network.

Clark and Mcfarland (1979) reported on cottonseed optical properties related to seed viability. The wavelength range used for the study was 720 - 800 nm. The results showed that optical transmittance of whole seed is correlated with seedcoat properties, which are important for seed germination. Otoni et al., (2008) used x-ray analysis to assess seed vigor of cotton. Cottonseed was classified into categories related to embryo size and the presence of damage on the seed coat based on analysis of x-ray images.

Objective

There is a desire to increase seed cotton packaging densities to increase efficiency in harvesting, transport, and storage. Based on the literature, it is hypothesized that compression level, moisture content, and storage time affect the quality of cottonseed. The objective of this research is to quantify the impacts of these factors, individually and in combination, on the quality of cottonseed.

MATERIALS AND METHODS

Sample Descriptions and Preparation

The experimental design included 360 total treatments of three replications each. The treatments consisted of two cotton varieties, three compression-density levels, three moisture-content levels, four storage-length levels, and five different bale locations. The cotton was grown in 2012 at Texas A&M AgriLife Research's farm in Burleson County, Texas (96.431685° W, 30.530911° N). The soil types in the field where the cotton was grown were Belk clay (BaA) 0 to 1 percent slope, Weswood silty clay (WwA) loam 0 to 1 percent slope, and Yahola fine sandy loam (YaB) 0 to 2 percent slope. The two varieties grown were Phytogen 499WRF (Dow AgroSciences) and Deltapine 0935B2RF (Delta and Pine Land Co.). The seed cotton was harvested with a John Deere 9970 cotton picker.

A 30-foot wagon was used to store the 1814 kg (4000 lbs) of cotton harvested, roughly equally divided between the two varieties. A plywood wall was inserted in the middle of the wagon to separate the two varieties, and the wagon was stored under an open-sided shed. The wagon was covered with two layers of tarpaulin to prevent water from intruding in the cotton. A general-use tarp was used as a first layer with a heavy-duty tarp used as a second layer. While the harvested cotton was protected from rain, it remained in equilibrium with ambient atmospheric conditions and tended to be approximately at 9% moisture content wet basis (MCWB). Moisture content was determined according to ASABE Standard 358.2 (ASABE Standards, 1988).

Humidity Chambers

To achieve the three cotton moisture levels needed for the experiment, relative humidity (RH) chambers were used. Two chambers with dimensions of 6 x 3 x 1.5 ft that can hold 48 dry pounds of cotton each were built (Figure 2). Saturated water-salt solutions enclosed in each sealed chamber were used to create the RH levels needed to produce known equilibrium moisture contents. Three relative humidity levels – 33, 53, and 75% RH – were determined to closely relate to the three moisture levels needed (5, 8, and 11% MCWB) for the experiment (Griffin Jr., 1974). Three salts solution (magnesium chloride, magnesium nitrate, and ammonium sulfate, with solubility levels of 167, 125, and 76 g per 100 mL of water, respectively) were used to produce the desired RH levels (Wexler and Hasegawa, 1954). The cotton was stored in a humidity chamber for one month to reach equilibrium.

Figure 2. Humidity chamber.

Miniature Bale Press

A hydraulic bale press was designed and built (Figures 3 and 4) to compress bales of seed cotton. To maintain the relative dimensions of a large cotton bale, the dimensions were set at 16.6 in. by 8.3 in. by 6.2 in.. Square 10-gauge steel tubing was used to minimize deflection. The chamber was built with the longest dimension vertical, and adequate headspace was provided for loading seed cotton. Once hand-fed and manually compressed cotton reached the top of the bale chamber, the hydraulic cylinder was used to compress the cotton, and bale length was measured. This process was repeated for each experimental unit (bale) until the pre-defined cotton weight was completely compressed and the tramper foot clamped into place.

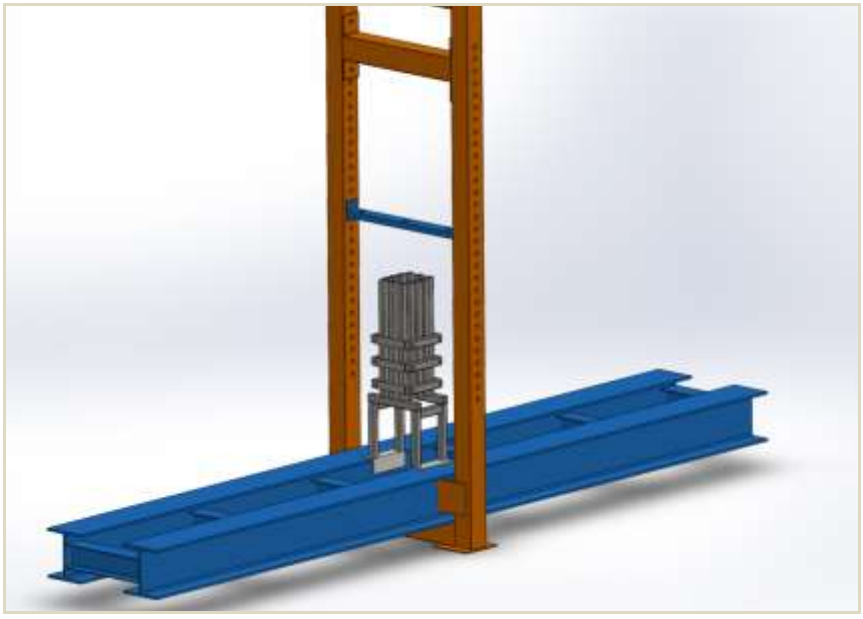

Figure 3. Solidworks drawing of bale press (Hartley, 2014).

One wall was then removed and two straps placed around the bale for binding. The bale was then weighed and its linear dimensions recorded with a measuring square in order to calculate the final volume of the bale. Load cells were used to measure the compression force in all directions. The analog signals were recorded by the data logger and were calibrated with a truck scale's digital reading.

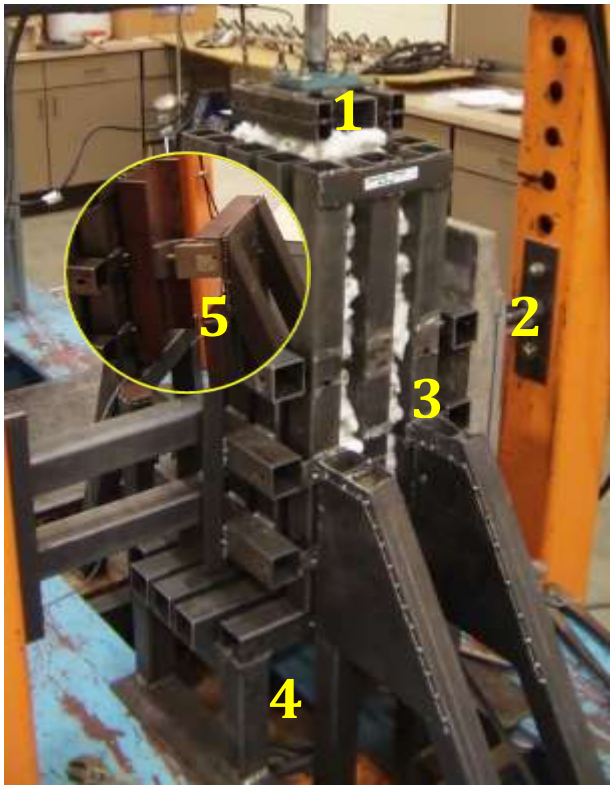

Figure 4. Miniature bale press showing the parts. Tramper foot (1), truck scale (2), bale chamber (3) and chamber stand (4). The inset in the circle shows the S-load cell at the back side of the chamber (5).

Bale Labelling and Ginning

Four samples from each bale were used, due to the possible variation in compression throughout the bale (Figure 5). Sampling locations are as follows:

1) ENDS: The two end faces in contact with tramper foot were combined.
2) NSM: Samples were collected to characterize the non-strapped side in contact with metal (non-strap metal) in order to quantify frictional damage at bale chamber walls due to shear stress during compaction.
3) NSNM: Samples were collected from cotton lying between side wall tubing (non-strap non-metal) in order to provide a comparison to metal contact.
4) 3INT: Three internal samples were collected from top, middle, and bottom of the bale and combined.

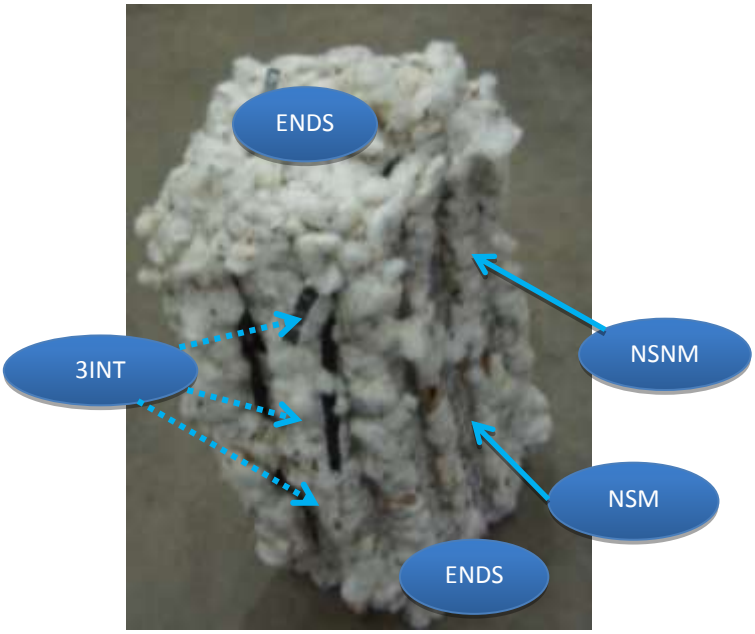

Figure 5. Sampling locations.

Samples were ginned at the Texas A&M Cotton Improvement Laboratory in College Station, Texas. Three Continental Eagle 10-saw gins (Continental Gin Co., Model Circa 1960) were used to gin all the samples. The seed from each sample were carefully collected and labeled for further analysis.

Cool Germination Test

Cool germination tests were carried out according to the criteria of the Association of Official Seed Analysts (AOSA), which require a temperature of 18°C for seven days. Each test was conducted with 50 non-delinted seeds from every sample locations in a bale, and an aggregate (AGG) was created with the combination of 12 seeds from each sample location: ENDS, NSM, NSNM and 3INT. A total of 51,840 cottonseed were included in the germination test. Seed germination paper (Anchor Paper Co., Model K-24) with a size of 25.4 cm x 50.8 cm (10 in. x 20 in.) was used, and the between-paper (BP) method was used for the germination test (ISTA, 1976).

For every sample, three germination papers were hand-wetted with purified water from a mist sprayer. The water was applied such that it would not be dripping from the paper (Savoy, 2005). Two attached wet papers were laid down, and the 50 seeds were evenly spaced out on them. The third paper was placed on top to cover the seeds, and then the entire assembly of papers and seeds was rolled carefully so that no seed would fall off. Each rolled assembly was placed inside an airtight plastic crisper (Pioneer Plastics, Model 395C) in upright position to allow for the drainage of excess moisture. When a crisper was full it was labelled and placed in a wooden compartment. The

compartment was covered with black duct-tape and black papers to ensure the inside was dark and no light could enter. A digital room thermometer was used to monitor the compartment's temperature, which was maintained at 18°C. The dates of placement and removal were recorded so the seven-day period could be tracked accurately. At the end of seven days, the paper assemblies were unrolled, and "normal germinated seedlings" were counted. Seedlings with a combined hypocotyl and radicle length of 3.81 cm (1.5 in) or longer were considered to have undergone normal germination (Figure 6).

Figure 6. Germination test.

Crackage Study and Counting Procedure

All seed used in the crackage study were delinted prior to evaluation. A handful of fuzzy seed from each sample and bale location was placed in a perforated container

which was placed in a bath of 93% concentrated sulfuric acid (H_2SO_4). A small piece of thick PVC with a number carved on it was placed in the container to identify the seed after the cleansing and drying had taken place. The numbers on the PVC piece and the aluminum pan were recorded so they could be matched after drying. The mixture of seeds and sulfuric acid was stirred continually with a wooden rod to ensure uniform contact between the delinting agent (H_2SO_4) and the lint.

The duration of the sulfuric acid treatment was about 2.5 to 3.0 minutes. At the end of that time, tap water was run freely over the seeds to wash the acid from their surfaces. The seeds then were removed from the container and placed into an aluminum pan with the PVC tag to maintain their sample numbering. The seeds were then placed in an oven for drying at 65°C (149°F) for at least 5 hours to remove any remaining moisture.

Dried delinted seeds were placed in Ziploc plastic bags and labelled to match their source samples. Each seed was examined and categorized (Table 2) as having no cracks (Category 1), having a crack (Category 2), or having part of the seed missing (Category 3). A control group of seeds were intentionally cracked (Figure 7a) by compressing them with a hand compression tool prior to delinting. This control group was used for visual comparison with seeds suspected as being cracked.

One hundred seeds from each sample bag were counted out onto a seed counting board (Figure 7b). Each seed was then manually picked up with tweezers, and all sides of the seed were visually inspected in detail to observe any damage on it. These seed coat inspections had to be conducted with rigorous attention to detail to ensure no

residual lint was mistaken for cracks or vice versa. Careful inspection also had to be made to ensure that multiple seeds in the "part missing" category were not from the same broken seed. When a final determination was made to categorize a seed, it was placed in the appropriate category pile for the given sample. The seeds in each pile were counted and the number recorded. Finally, the counted seeds were placed in a separate bag and labelled to match their source samples, but this bag had the "CA" designation added at the end to indicate crack analysis. Enlarged pictures of the seed damage are shown in Figure 8.

(a) **(b)**

Figure 7. (a) Intentionally cracked seeds; (b) Counting board.

Table 2. Seed damage classifications and its descriptions.

Category	Type of damage	Descriptions
1	No crack	Seed has no visible crack.
2	Cracked	Seed is visibly cracked, but no part appears to be missing.
3	Part missing	Part of the seed is missing due to excessive damage.

a) 2nd category

b) 3rd category

Figure 8. Enlarged pictures of the cracked seeds.

Data Analysis

The effects of seed cotton compression on incomplete germination and crackage of cottonseed, accounting for interactions among varieties, densities, moisture contents, storage times, and locations of the sample in the bale, were analyzed with the Design-Expert statistics package (version 9.0.0.7, Stat-Ease Inc.). The software allows the experiment to have categorical and numerical factors with multiple levels. Furthermore,

all the possible combinations and interactions between the independent variables and dependent variables could be found with the statistical design. Two-dimensional and three-dimensional contour plots were drawn to evaluate the interaction of independent variables with the chosen dependent variables.

Instead of using nominal values of density and moisture content, actual measured values were used to improve the analysis and better reflect experimental results. Variety and location factors were specified as categorical with a nominal subtype, while density, moisture, and storage were specified as numerical with a discrete subtype (Table 3). A Multilevel Factorial Design was used to construct a linear regression with interaction among the five design factors and two optimization parameters (responses). The design consisted of 360 trials for each response, replicated three times each, thus giving 1080 total runs.

Table 3. Variables name and types.

Factor	Name	Units	Type	Subtype
A	Variety (2 levels)		Categorical	Nominal
B	Density	lb ft^{-3}	Numerical	Discrete
C	Moisture	%	Numerical	Discrete
D	Storage	days	Numerical	Discrete
E	Location (5 levels)		Categorical	Nominal

The Design-Expert software was used to determine the various components of the full-model Analysis of Variance (ANOVA). Model reduction was used to simplify a regression model by eliminating insignificant terms. Reducing the number of terms could make the model less complicated and easier to work with, and insignificant terms left in the model could reduce the precision of the predictors. A model was reduced by determining which terms were statistically significant by examining the p-value of each coefficient. The coefficients with p-values greater than 0.05 were removed, and those with p-values less than 0.05 were maintained.

All the hierarchical, quadratic and cubic relationships among factors were not tested because of their added complexity with doubtful benefit, so only two-factor interaction terms were included. The Tukey HSD (honest significant difference) post-hoc test was used to determine which groups in the treatments differed from each other when significant differences were found by ANOVA.

RESULTS AND DISCUSSION

Table 4 summarizes the data from the incomplete germination and crackage studies. The incomplete germination value for a given sample indicates the portion of the seeds that failed to germinate. The mean incomplete germination value across all samples was approximately 15%. The crackage value for a given sample indicates the portion of the seeds that had identifiable cracks. The mean crackage value across all samples was approximately 9%. The minimum percentage for incomplete germination was 1% and the minimum percentage for crackage was 0.5%. The maximum percentage and standard deviation for incomplete germination and crackage studies were about the same, 56% and 8%, respectively.

Table 4. Response summary.

Response	Name	Minimum	Maximum	Mean	Std. Dev.
R1	Incomplete Germination	0.01	0.56	0.1549	0.0806
R2	Crackage	0.005	0.56	0.0936	0.0817

The analysis of variance (ANOVA) results of the incomplete germination study for full and reduced models are shown in Tables 5 and 6, respectively. Estimates made with the full model were significantly correlated to the actual data ($p < 0.0001$), but the amount of incomplete germination variability was not well explained by the variability in the model parameters ($R^2 = 0.153$). Several main effects were significant at the 5%

level in the full model and were thus included in the reduced model: B (density), C (moisture), D (storage), E (location) and interaction effects BC (density-moisture), BD (density-storage), BE (density-location), and CD (moisture-storage). Estimates made with the reduced model were also significantly correlated to the actual data ($p < 0.0001$, $R^2 = 0.143$).

Table 5. **ANOVA full model for germination test.**

Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F
Model	1.08	30	0.036	6.33	< 0.0001
A-Variety	0.010	1	0.010	1.84	0.1749
B-Density	0.13	1	0.13	22.27	< 0.0001
C-Moisture	0.32	1	0.32	56.74	< 0.0001
D-Storage	0.057	1	0.057	10.10	0.0015
E-Location	0.13	4	0.031	5.53	0.0002
AB	0.00156	1	0.00156	0.28	0.5989
AC	0.00293	1	0.00293	0.52	0.4723
AD	0.00748	1	0.00748	1.32	0.2507
AE	0.00744	4	0.00186	0.33	0.8587
BC	0.026	1	0.026	4.66	0.0311
BD	0.065	1	0.065	11.44	0.0007
BE	0.13	4	0.033	5.85	0.0001
CD	0.15	1	0.15	25.86	< 0.0001
CE	0.00313	4	0.00078	0.14	0.9682
DE	0.042	4	0.010	1.85	0.1174
Residual	5.94	1049	0.00566		
Cor Total	7.01	1079			
Std. Dev.	0.075				
Mean	0.15				
C.V. %	48.59				
R-squared	0.1534				
Pred R-quared	0.0974				

Table 6. ANOVA reduced model for germination test.

Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F
Model	1.00	14	0.072	12.67	< 0.0001
B-Density	0.13	1	0.13	22.33	< 0.0001
C-Moisture	0.31	1	0.31	55.60	< 0.0001
D-Storage	0.055	1	0.055	9.68	0.0019
E-Location	0.13	4	0.031	5.54	0.0002
BC	0.027	1	0.027	4.81	0.0284
BD	0.064	1	0.064	11.41	0.0008
BE	0.14	4	0.035	6.19	< 0.0001
CD	0.17	1	0.17	29.26	< 0.0001
Residual	6.01	1065	0.00565		
Cor Total	7.01	1079			
Std. Dev.	0.075				
Mean	0.15				
C.V. %	48.52				
R-squared	0.1428				
Pred R-quared	0.1165				

The final equation in terms of coded factors for the incomplete germination is as follows:

$$Y_1 = 0.015 + 0.016B - 0.014C - 0.011D - 0.0034D + 0.010E\,[1] + 0.010E\,[2] - 0.010E\,[3] - 0.0097E\,[4] + 0.0087BC + 0.013BD - 0.0024BE\,[1] + 0.006BE\,[2] - 0.011BE\,[3] - 0.017\,BE\,[4] + 0.021CE\ ;$$

Where Y_1 - Percent of incomplete germination (%)

B - Compression density (lb ft^{-3})

C - Moisture contents (%)

D - Storage times (days)

E - Locations of the sample in the bale

In the analysis of crackage, the full model was not only significant ($p < 0.0001$), but it also had an R^2 value of 0.64, indicating that the model accounts for 64% of data

variability. The full model summary is shown in Table 7, and the reduced model (including only significant factors in the full model) is shown in Table 8. Crackage was affected by the B (density, $p < 0.0001$), C (moisture, $p < 0.0259$), D (storage, $p < 0.0001$), E (bale location, $p < 0.0001$), the interaction of B and D (density-storage, $p < 0.0001$), and the interaction of B and E (density-location, $p < 0.0001$).

Table 7. ANOVA full model for crackage study.

Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F
Model	4.64	30	0.15	63.53	< 0.0001
A-Variety	0.00152	1	1.522E-003	0.62	0.4295
B-Density	2.82	1	2.82	1155.32	< 0.0001
C-Moisture	0.012	1	0.012	4.84	0.0280
D-Storage	0.040	1	0.040	16.29	< 0.0001
E-Location	0.84	4	0.21	85.97	< 0.0001
AB	0.00110	1	0.00109	0.45	0.5037
AC	0.00553	1	0.00552	2.27	0.1323
AD	0.00014	1	0.00014	0.059	0.8083
AE	0.00217	4	0.00054	0.22	0.9259
BC	0.00014	1	0.00014	0.059	0.8084
BD	0.044	1	0.044	18.15	< 0.0001
BE	0.83	4	0.21	84.67	< 0.0001
CD	0.0003	1	0.00030	0.12	0.7255
CE	0.00319	4	0.00080	0.33	0.8598
DE	0.011	4	0.00277	1.14	0.3369
Residual	2.56	1049	0.00244		
Cor Total	7.20	1079			
Std. Dev.	0.049				
Mean	0.094				
C.V. %	52.75				
R-squared	0.6450				
Pred R-squared	0.6229				

Table 8. ANOVA reduced model for crackage study.

Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F
Model	4.62	12	0.38	159.03	< 0.0001
B-Density	2.82	1	2.82	1163.44	< 0.0001
C-Moisture	0.012	1	0.012	4.98	0.0259
D-Storage	0.037	1	0.037	15.43	< 0.0001
E-Location	0.84	4	0.21	86.56	< 0.0001
BD	0.045	1	0.045	18.76	< 0.0001
BE	0.83	4	0.21	86.21	< 0.0001
Residual	2.58	1067	0.00242		
Cor Total	7.20	1079			
Std. Dev.	0.049				
Mean	0.094				
C.V. %	52.57				
R-squared	0.6414				
Pred R-squared	0.6321				

The final equation in terms of coded factors for the crackage is as follows:

$$Y_2 = 0.091 + 0.068B - 0.0043C + 0.0066D + 0.022E\,[1] + 0.030E\,[2] - 0.023E\,[3] - 0.035E\,[4] + 0.010BD + 0.0175BE\,[1] + 0.045BE\,[2] - 0.033BE\,[3] - 0.046BE\,[4] - 0.017BC\,[4] + 0.021CD\ ;$$

Where Y_2 - Percent of crackage damage (%)
B - Compression density (lb ft^{-3})
C - Moisture contents (%)
D - Storage times (days)
E - Locations of the sample in the bale

Figures 9a and 9b show surface plots of the effects on germination by the interaction between storage and density at the 5% and 11% moisture levels, respectively. Since variety was not a major factor, only the Phytogen 499 variety and AGG

(aggregate) as a representative bale location were used to produce these plots. At longer storage times, incomplete germination tended to be greater as density increased, regardless of moisture content, but the effect was pronounced at higher moisture content. At 5% moisture, incomplete germination increased from 11 to 15% as density increased from 18 to 30 lb ft^{-3} (pcf), while at 11% moisture, it increased from 11 percent 18 percent – almost twice the increase. It is plausible that increased moisture enabled more biological activity during storage, and that increasing compression density produced cracks in the seed that enabled this biological activity to increase damage to the seed. At shorter storage times the change in incomplete germination was minimal. Even though there were interactions between moisture content and other factors, there was no significant difference in the incomplete germination values between different levels of moisture content (5, 8 and 11%) as shown by the Tukey HSD test (Table 9).

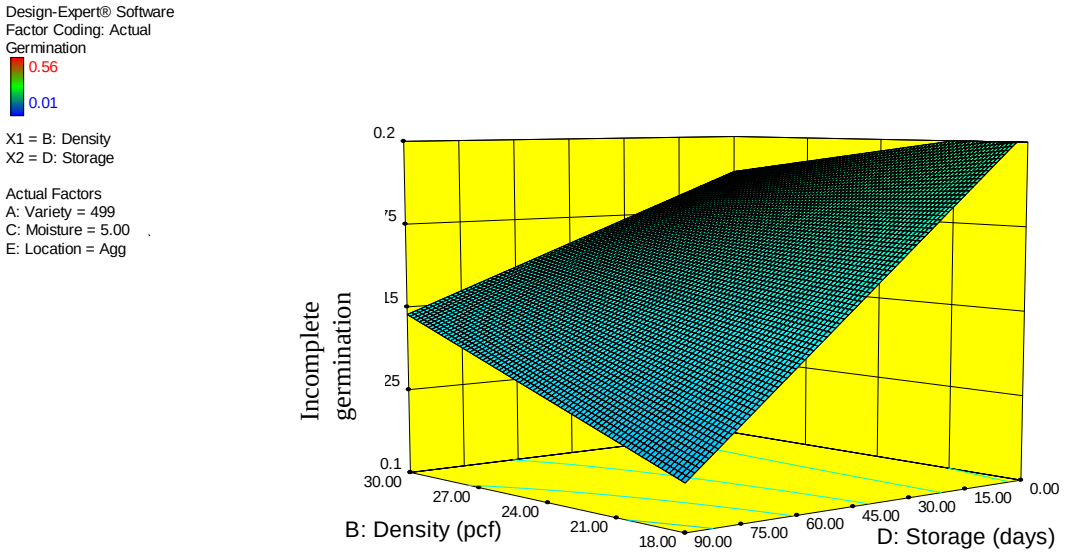

Figure 9a. 3D surface plots of incomplete germination-storage-density interaction at 5% moisture content.

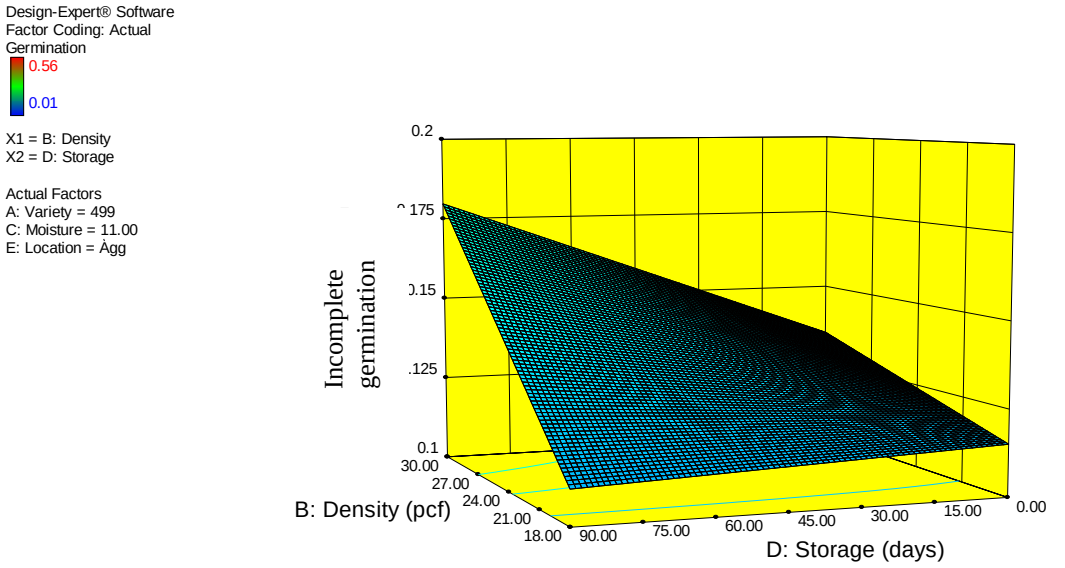

Figure 9b. 3D surface plots of incomplete germination-storage-density interaction at 11% moisture content.

Table 9. Tukey's mean comparison of incomplete germination for moisture content.

Moisture content (%)	Mean
8	0.0967 [A]
5	0.0948 [A]
11	0.0892 [A]

*Levels not connected by same letter are significantly different

Figure 10 shows a graph of crackage vs. storage time at two compression densities (18 and 30 lb ft^{-3}). At the lower density, crackage was not influenced by storage time, but at the higher density, crackage increased markedly with the storage time. The Tukey post-hoc test showed that the means of crackage were significantly different among all levels of compression density (Table 10). In addition, the mean value of crackage damage for 30 lb ft^{-3} increased to more than twice the mean value of crackage at 24 lb ft^{-3}. As regards germination, however, only the mean of incomplete germination at the highest density (30 lb ft^{-3}) was different from the other means (Table 11). Considering these trends together, it is likely that increasing compression density resulted in increased crackage, and extended storage time particularly at higher moisture levels exacerbated seed damage through biological activity, ultimately reducing germination.

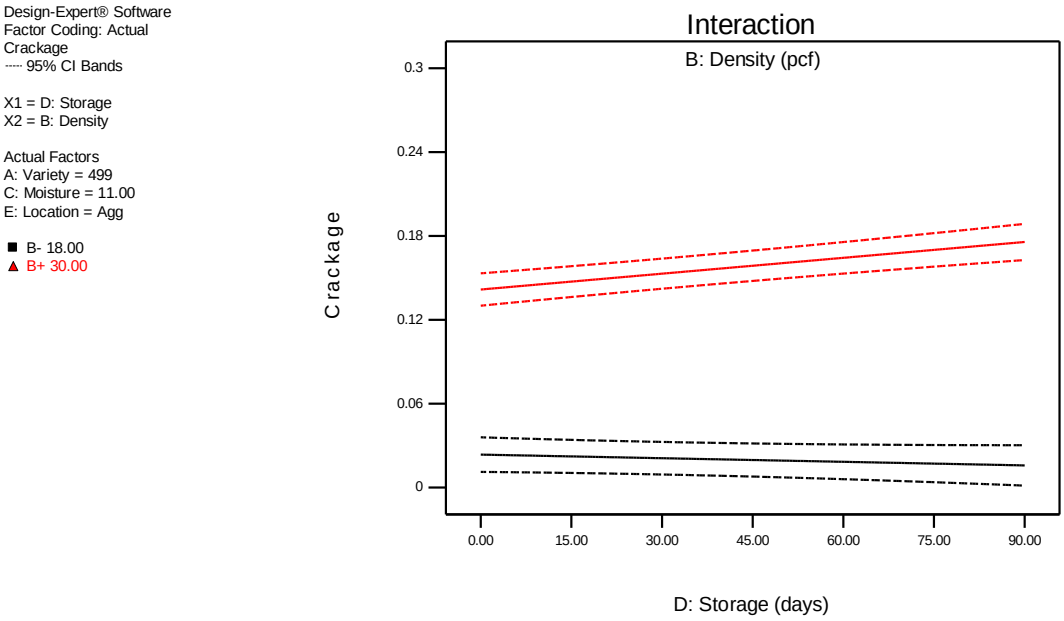

Figure 10. **Graph of crackage-storage interaction in different density.**

Table 10. Tukey's mean comparison of crackage damage for compression density.

Compression density (lb ft^{-3})	Mean
30	0.1700 ^{A}
24	0.0671 ^{B}
18	0.0438 ^{C}

*Levels not connected by same letter are significantly different

Table 11. Tukey's mean comparison of incomplete germination for compression density.

Compression density (lb ft^{-3})	Mean
30	0.1777 [A]
18	0.1487 [B]
24	0.1381 [B]

*Levels not connected by same letter are significantly different

Figure 11 shows the percentage of incomplete germination at different moisture contents and storage times. It can be seen that at the minimum storage time (0 days), incomplete germination decreased as moisture content increased. However, at the maximum storage time (90 days) increasing moisture content was related to an increase in incomplete germination. It is possible that higher moisture contents reduced the propensity of the seed to crack, a trend that would show up as reduced incomplete germination with higher moisture content when storage was not a factor. It is also possible that a lengthy storage time could reverse the effects of such a trend, in that higher moisture content would be the overriding factor in increasing damage due to biological activity regardless of the presence of cracks. The Tukey HSD comparison at 0.05 significance level indicated that the mean of incomplete germination at 0 storage time was significantly higher than at the other three storage times (7, 30, and 90 days), but there was no difference in incomplete germination among the non-zero storage times (Table 12). It is possible that, particularly at low starting moisture contents, the seed took up moisture during storage, and the initially dry state of the seed may have caused the

mean of incomplete germination at 0 storage time to be higher than the other three storage times.

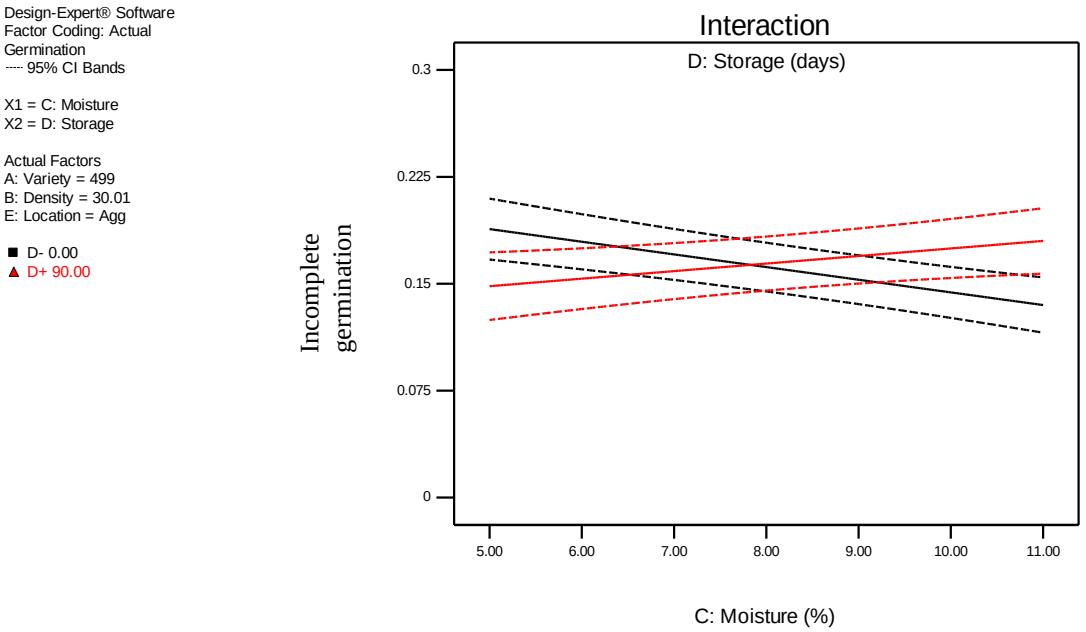

Figure 11. Graph of incomplete germination-moisture interaction in different storage time.

Table 12. Tukey's mean comparison of incomplete germination for storage length.

Storage time (day)	Mean
0	0.1738 ^{A}
7	0.1520 ^{B}
90	0.1513 ^{B}
30	0.1424 ^{B}

*Levels not connected by same letter are significantly different

Figure 12 shows crackage at different compression densities and storage times. It is apparent that crackage was linearly related to compression density and that storage time had little effect. However, the Tukey post-hoc comparison indicated that mean crackage at 90 days of storage was significantly higher than at lesser storage times (Table 13). The storage-time effect appeared to be small but became more apparent as interactions with density and moisture were considered (Figures 13a through 13e).

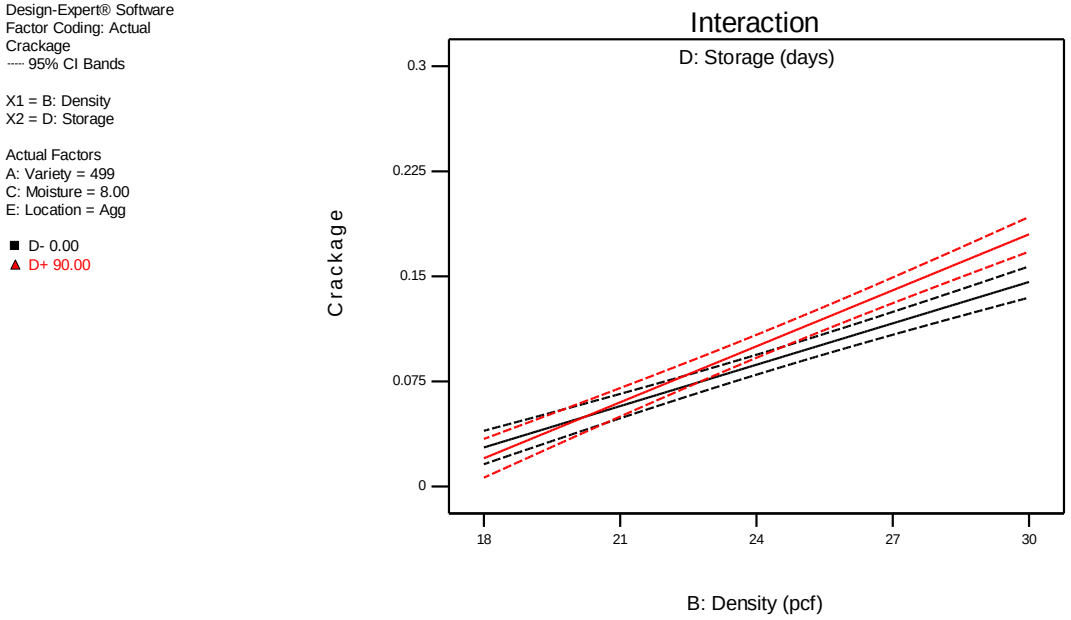

Figure 12. Graph of crackage-density interaction in different storage time.

Table 13. Tukey's mean comparison of crackage damage for storage length.

Storage time (day)	Mean
90 days	0.1097 [A]
7 days	0.0893 [B]
0 day	0.0878 [B]
30 days	0.0875 [B]

* Levels not connected by same letter are significantly different

To look carefully at the effects of compression density and storage time on crackage, the Phytogen 499 variety and 8% moisture level were used in the surface plots of Figures 13a through 13e. Moisture content was a significant factor, but its p-value of 0.0259 indicated a weaker relationship than with density and storage. Thus, 8% moisture was chosen as a moderate and representative level.

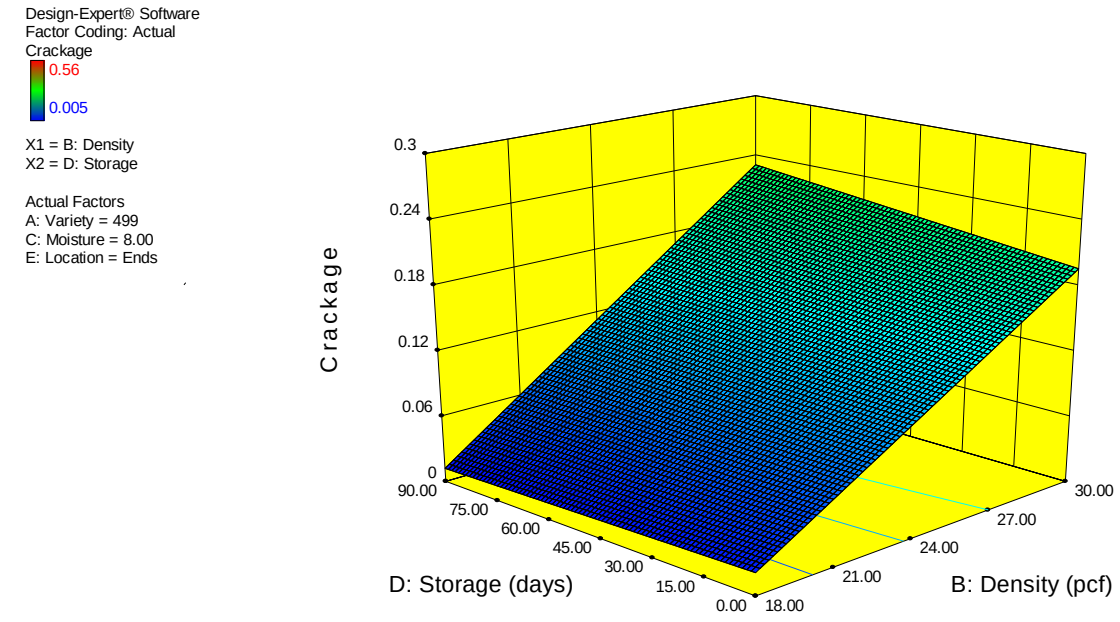

Figure 13a. 3D surface plots of crackage-density-storage interaction in ENDS location.

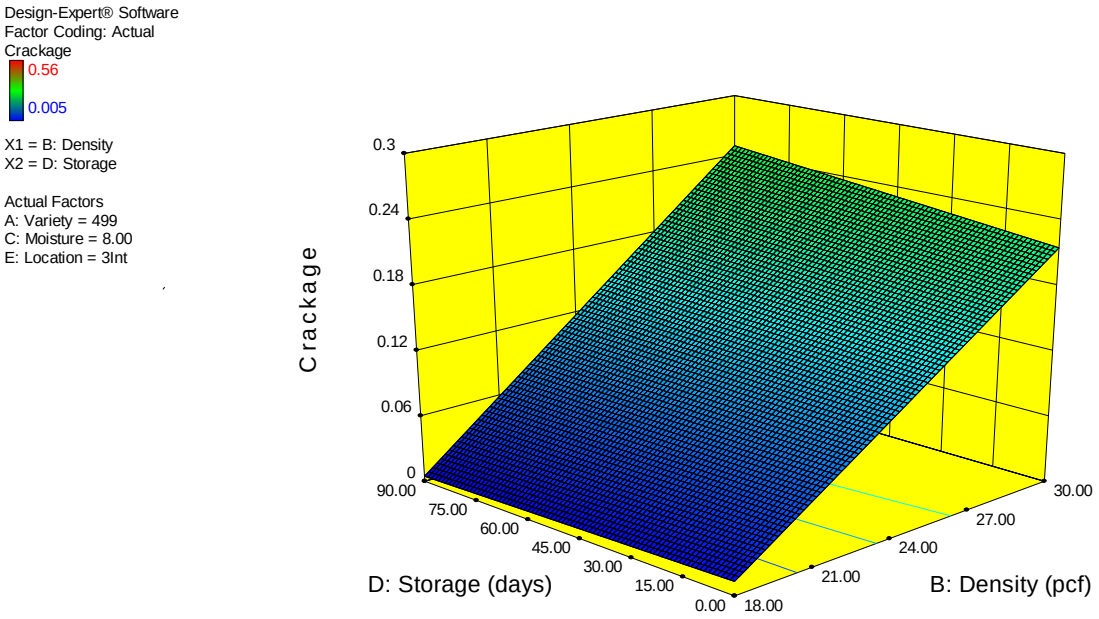

Figure 13b. 3D surface plots of crackage-density-storage interaction in 3INT location.

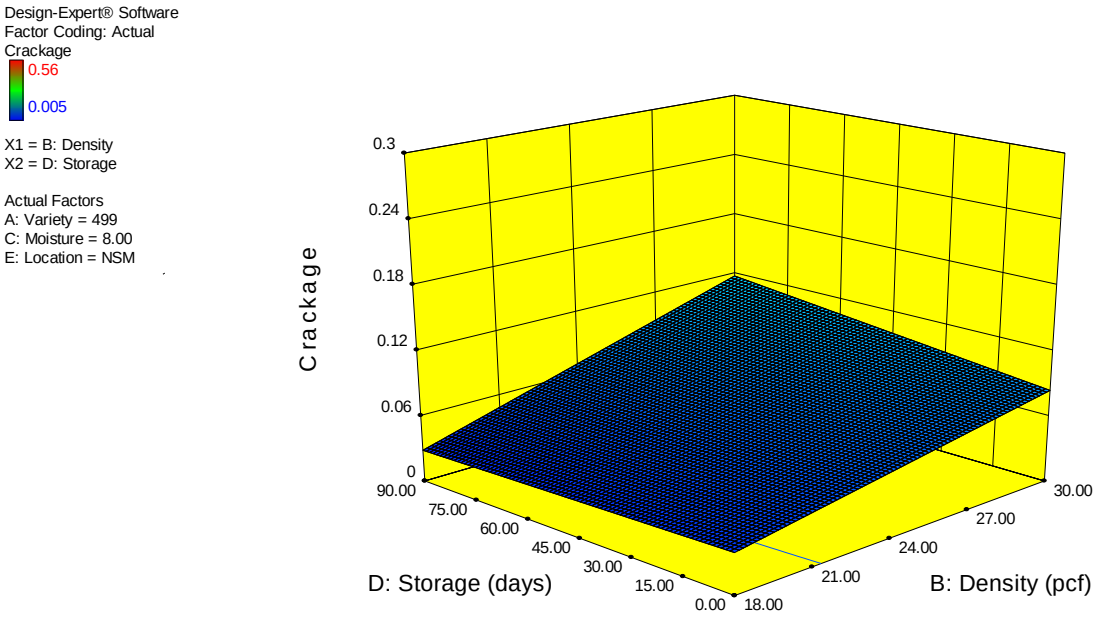

Figure 13c. 3D surface plots of crackage-density-storage interaction in NSM location.

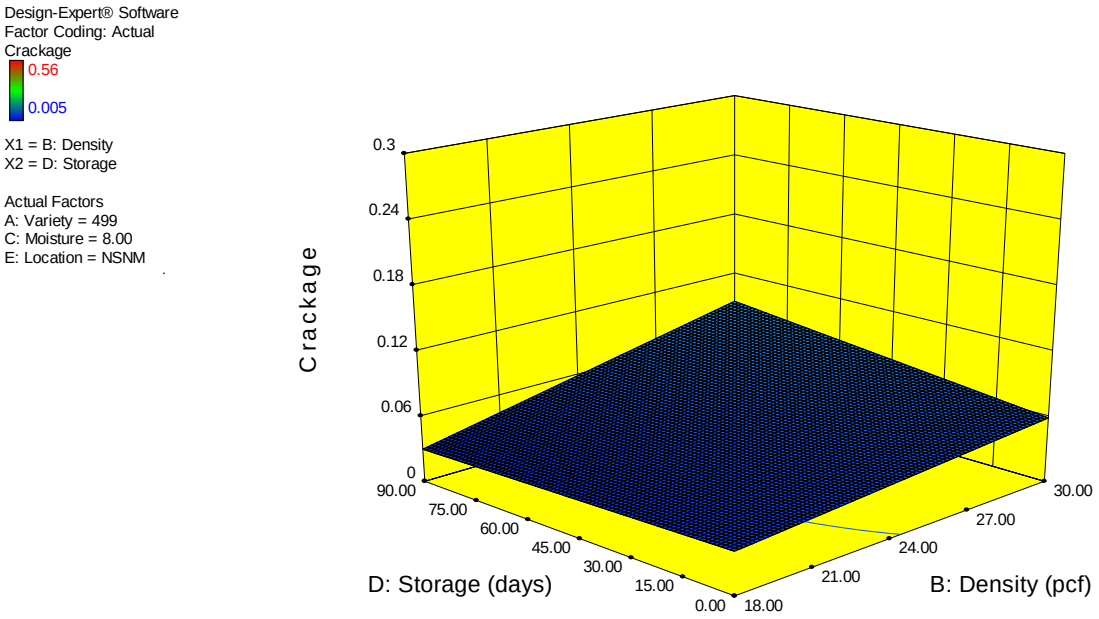

Figure 13d. 3D surface plots of crackage-density-storage interaction in NSNM location.

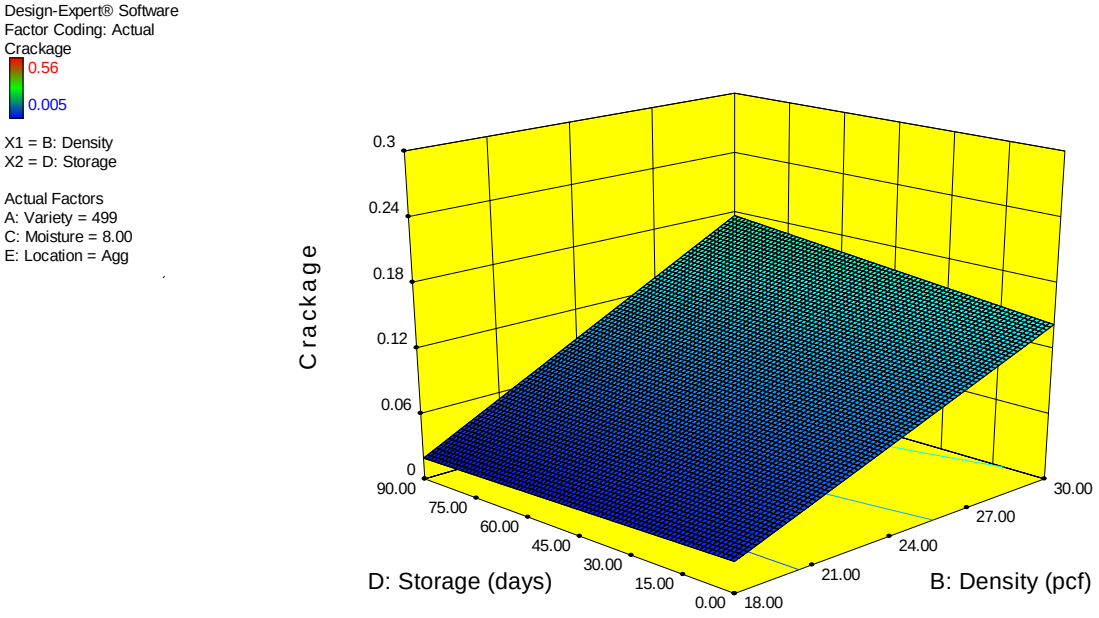

Figure 13e. 3D surface plots of crackage-density-storage interaction in AGG location.

It is clear in these figures that crackage increased with compression density. A clear picture of the relationship is shown in the least significance difference (LSD) graph of Figure 14, in which the crackage difference between 18 and 30 pcf compression density is pronounced.

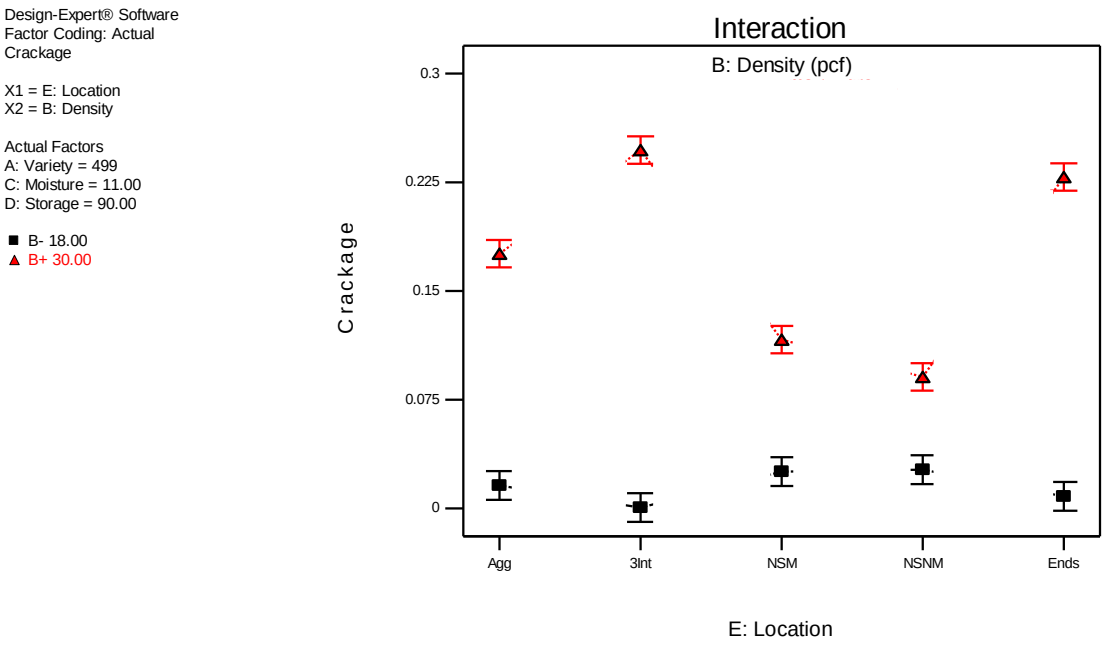

Figure 14. Graph of Least Significance Difference (LSD) crackage damage for 18 and 30 pcf.

Crackage was higher in the ENDS and 3INT locations than in the NSM and NSNM locations (Table 15). The mean value for crackage in ENDS and 3INT locations was almost twice that of NSM and NSNM. The fact that crackage was higher in ENDS may be because the seed had been in contact with or very close to the tramper foot and walls, which may have been areas of particularly high stress. In 3INT, the reason may be

because of the level of bulk stress was highest in the middle of the bale. There was no significant difference between the crackage and germination values in NSM (in contact with metal at the sides) and NSNM (not in contact with metal at the sides), as shown by Tukey HSD test (Tables 14 and 15).

Table 14. Tukey's mean comparison of crackage damage for location.

Location	Mean
Ends	0.1268 [A]
3Int	0.1205 [A]
Agg	0.0959 [B]
NSM	0.0688 [C]
NSNM	0.0558 [C]

*Levels not connected by same letter are significantly different.

Table 15. Tukey's mean comparison of incomplete germination for location.

Location	Mean
Ends	0.1695 [A]
3Int	0.1656 [A]
Agg	0.1513 [AB]
NSNM	0.1440 [B]
NSM	0.1439 [B]

*Levels not connected by same letter are significantly different.

Comparing the results between the germination and the crackage studies, the same four effects are significant at the 5% level: B (density), C (moisture), D (storage) and E (bale location). Two effect interactions are also significant at the 5% level for both studies: BD (density-storage) and BE (density-location). In the germination study, the

BC (density-moisture) interaction was also significant, but the p-value was closer to the 5% limit ($p = 0.0284$) than for the other effects and interactions. In the crackage study this interaction effect was not significant. While moisture was significant in the crackage study, it was closer to the 5% limit ($p = 0.0259$) than the other effects.

In the germination study, moisture had the highest F value (55.6), followed by density (22.33), storage time (9.68) and bale location (5.54). It was found that the highest incomplete germination occurred in the presence of high moisture content. Since moisture is a dominant factor in the quality of stored cottonseed, it stands to reason that when cottonseed are stored in a high-density seed-cotton package with minimal ventilation, the likelihood that they will lose their viability should increase at higher moisture levels.

In the crackage study, density had the highest F value (1163.44), followed by bale location (86.56), storage time (15.43), and moisture (4.98). Increasing compression density was associated with increased crackage, and certain bale locations where compression could be expected to be higher had increased crackage. It appears that compression densities achieved in this experiment exceeded seed coat strength in many circumstances. Moisture was not found to be strongly associated with crackage. In other studies (Comer, 1968; Brashears et al., 1970; Alemayehu, 1984) higher crackage at lower moisture contents may have been due to increased brittleness in the seed coat caused by lower moisture.

CONCLUSIONS

Two tests were used to evaluate the compression damage undergone by cottonseed during bale-type compression, germination and crackage. Results indicate that compression densities, moisture contents, storage times and locations of the sample in the bale were correlated with seed damage. Moisture was the most prevalent factor related to germination percentage followed by compression density, storage time and location. Increased moisture during storage was associated with incomplete germination, a response that could be expected as increased moisture encourages biological activity and brings about higher free fatty acid content, which causes deterioration of the seed. Compression density was the major factor in increasing crackage, followed by bale location, storage time, and moisture.

The variety of seed cotton used in the compression test was not significant in determining seed damage. Interactions between compression density and moisture content, compression density and storage time, compression density and bale location, and moisture content and storage time were significant in germination reduction. As storage time increased, the relationship between moisture content and germination percentage became more prominent. Increases of storage time and compression density also appeared to negatively impact germination.

Interactions between compression density and storage time, and compression density and bale location, were significantly related to crackage. As the storage time increased, the relationship between compression density and crackage became more

prominent. Bale location was also significantly related to crackage, as higher crackage was observed at the end and interior locations of the bale than at the side locations.

The results of this research are potentially important to cotton producers and the seed industry. If high-density compression begins to be used during harvesting to provide more efficient field packages for transport to the gin, seed cotton moisture contents will become even more critical to minimize seed damage. The maximum safe seed moisture content for storage is 12% (Lalor et al., 1995; Wilkes, 1974). Furthermore, high storage time at high density and moisture content could pose problems as well. The most striking result, which resembles the prior report of Brashears et al. (1970), suggests that compression density should not exceed 24 lb ft^{-3} to avoid significant damage to the cottonseed.

Further research to improve compression density is important to minimize seed cotton bale or module size for transport and storage. In future research, it may be useful to consider the effects of temperature and trash content. The literature suggests that effects of the temperature associated with moisture will influence how long the cotton can be stored. Also, the presence of foreign matter like burrs, sticks and leaves can add moisture to the seed cotton during storage. Finally, an optoelectronic sensing system could potentially be developed for automated seed damage detection. Such a system would reduce experiment time and might even provide higher accuracy in measuring cracks in large sample seeds.

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