Effect of Crop Seed Water Content on the Rate of Seed Damage by Red Imported Fire Ants (Hymenoptera: Formicidae)

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ABSTRACT Red imported fire ants, *Solenopsis invicta* Buren, are known to feed upon planted field crop seed; however, the relationship with varying levels of seed water content is not known. If red imported fire ants are feeding on planted seed during periods of slow seed germination, and if the seed are more palatable or otherwise more easily damaged at elevated water contents, the risk of crop-stand loss is increased. In this study, 5 types of field crop seeds—wheat, *Triticum aestivum* L.; corn, *Zea mays* L.; grain sorghum, *Sorghum bicolor* (L.) Moench; cotton, *Gossypium hirsutum* L.; and soybean, *Glycine max* L.—were exposed to red imported fire ants under laboratory conditions to measure damage caused by feeding. The seeds were dry and on moistened filter paper for maximum rate of germination, and at 20, 40, and 60% water contents for various parts of the study. Moistened seed suffered 2-90 times more damage in 48 h than comparable dry seed. When commercial sorghum seed was tested at 4 initial water contents, seed damage increased ≈22 times over damage to dry seed. A model was developed to predict the feeding damage to commercial sorghum seed as a function of elapsed time and seed water content. The risk of feeding damage relative to germination increased up to as much as 20 times when the initial water content was 60% compared with dry seed, except for corn damage. which was relatively insensitive to water content. We conclude from laboratory studies that crop-stand losses are likely to be most severe for wheat and grain sorghum when moistened seed are exposed to feeding by red imported fire ants.

KEY WORDS *Solenopsis invicta*, corn, wheat, sorghum, soybean, cotton

RED IMPORTED FIRE ants, *Solenopsis invicta* Buren, have been found to feed on seed of major crops grown in the infested region of the United States (Drees et al. 1991, 1992). Morrison et al. (1997) reported the rates of red imported fire ant feeding on the dry seed of wheat, *Triticum aestivum* L.; corn, *Zea mays* L.; grain sorghum, *Sorghum bicolor* (L.) Moench; cotton *Gossypium hirsutum* L.; and soybean, *Glycine max* (L.) Merr. They found that stored product insecticides used for commercial seed reduced the feeding damage rate by 54-94%. For example, feeding damage to corn seed was reduced from 6.44 to 0.88%/d, which is a decrease of 86%. Feeding damage was for dry seed without any indication of the effect of increased seed water contents, which occur naturally during water imbibition, germination, and emergence in moist soil. Fincher (1989) reviewed the changing chemical properties of cereal seed during the transformation from dormancy to germination. The effects of hydrolytic enzymes and other reported products of germination on red imported fire ant feeding are unknown. Drees et al. (1991) found that red imported fire ant feeding damage increased after soaking both corn and sorghum seed in water for 24 h. Their results indicate a dynamic situation in which higher water content induces faster seed germination as well as more aggressive feeding by red imported fire ants. Thus, as time elapses after planting, there is a risk that successful plant establishment cannot occur before red imported fire ant damage occurs.

Seed feeding by red imported fire ants can be affected by commercial seed treatments, colony size, and hunger status. Drees et al. (1992) and Collins (1996) demonstrated that seed protectants do not repel ants, but rather offer seed protection by preventing recruitment of additional ants to the treated material. Any effects of commercially applied stored product insecticides on the control of feeding damage are not known in relation to seed water content. Colony size was found to be positively correlated with seed-feeding damage during limited exposure periods in laboratory studies (Drees et al. 1991). They also found that seed-feeding can be altered by depriving the ants of food before the tests. Therefore, for tests, colonies should be approximately equal in size and treated uniformly to equilibrate the hunger status among colonies. Percentage seed germination can be used as the index of seed damage, as shown to be appropriate for dry seed by Morrison et al. (1997).

The objective of our study was to document rates of red imported fire ant feeding damage on seeds of wheat, corn, grain sorghum, cotton, and soybean when seed water content varied between dry and that

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Table 1. Seed type, insecticide, fungicide, and/or inoculation coating, variety, and sizing sieve

Seed treatment	Coating[a]	Variety	Sieve size
Wheat (n)	None	McGregor 2180[b]	#8
Corn (n)	None	DeKalb KD-656[b]	6.4 mm
Corn (c)	Captan and Actellic	DeKalb DK-656[b]	6.4 mm
Sorghum (n)	None	DeKalb DK-37[c]	#6
Sorghum (c)	Captan and Reldan	DeKalb DK-37[c]	#6
Cotton (n)	None	GP 74+[d]	#4
Cotton (c)	Thiram and Apron	GP 74+[d]	#4
Soybean (n)	None	DeKalb CX-458[c]	#4
Soybean (c)	Soybean S culture	DeKalb CX-458[c]	#4

[a] Coatings were supplied-applied at 1994 manufacturer's recommended rates. Materials were as follows: (1) Captan (Dicarboximide), protectant–eradicant fungicide; (2) Thiram (Carboxin), seed protectant fungicide and animal repellent; (3) Apron (Metalaxyl), protectant fungicide; (4) Actellic (Pirimmiphos-Methyl), stored product insecticide; (5) Reldan (Chlorpyrifos-Methyl), organophosphate insecticide; (6) Soybean S culture, Nitaragin, legume innoculation culture, in slurry with Coca-Cola.

[b] ESCO Seed, McGregor, TX.

[c] DeKalb-Pfizer Genetics, Illiopolis, IL.

[d] G. & P. Seed, Aquilla, TX.

needed for maximum germination rate. The results may be used to estimate the risk of red imported fire ant damage to planted crop seeds in soils of various water contents.

Materials and Methods

The study was conducted at the Grassland Soil and Water Research Laboratory, Natural Resources Conservation Research Unit, USDA–ARS, Temple, TX, in cooperation with the Center of Medical and Veterinary Entomology, Imported Fire Ant and Household Insects Research Unit, USDA–ARS, Gainesville, FL.

Colonies of red imported fire ants were dug from reduced-tillage field areas, separated from the soil, and maintained in trays with artificial nests, water, and food (dead crickets and 1:1 dilution of honey and water) as reported by Morrison et al. (1997) and described by Banks et al. (1981) and Williams (1989). The colonies were maintained in a windowless laboratory, with temperature controlled at 25°C and 12 h of artificial light per day.

Tested crop seeds were from commercially available sources as both untreated, "natural," and commercially treated with stored product insecticides (coated) (Table 1). Soybean seed which is not sold with insecticidal coatings was tested both as natural seed and as inoculated seed (Soybean "S" Culture, Nitragin, Milwaukee, WI), with the inoculum suspended in a solution of sucrose-sweetened drinking soda (Coca-Cola; Coca-Cola, Atlanta, GA). Wheat was used as natural seed only in accordance with local planting practice.

Feeding trials were conducted in separate trays, each equipped with a watered colony cell, a watering tube, and a feeding dish (Morrison et al. 1997). The feeding dish was a polystyrene petri dish (100 mm diameter, 15 mm deep) with four 3.2-mm holes drilled into both the sides [illegible] prevent removal of whole seed. Seed for the study treatments was put into the feeding dish in each tray at the start of a trial, as detailed below. Red imported fire ant colonies were deprived of food for 5 d and subdivided into 2,000 workers and some brood in each tray.

Study procedures were separated into those for maximum seed germination rate to minimize seed damage, and those for seed maintained at subgermination water contents.

Maximum Rate of Seed Germination. This experiment was designed to document the rates of feeding damage to seed which were at the least risk to damage because of maximum germination rates. Individual lots of 50 dry seed were placed on wet filter paper in the feeding dishes for all 9 combinations of seed type and coatings (Table 1). Germination was counted at 24, 48, 72, and 96 h. Visually assessment of seed with damage (visible scars or greater damage) were counted at 3, 7, 24, 48, 72, and 96 h and converted to percentage of the 50 seeds which were damaged. Seed water content was monitored by weighing whole seed each time they were inspected for damage. There were 4 replications, each using red imported fire ants from different colonies.

Four Levels of Sorghum Seed Water Content. This experiment was designed to establish sorghum seed damage rates at discrete water contents for the development of a model. Coated grain sorghum seeds were studied at dry (seed water content was in equilibrium with ambient air humidity) and premoistened water contents of ≈20, 40, and 60% dry basis. For the premoistened conditions, water was added to dry seed, followed by refrigeration for 4 d for moisture equilibration without germination. Five replications were conducted simultaneously; a 6th replication was not exposed to ant feeding but weighed to monitor seed water loss during the trial. Red imported fire ants from 5 colonies were used for the 5 replications. Seed damaged by feeding were counted at 3, 7, 24, 48, 72, and 96 h. Seed germination was not studied. The trial was repeated for a total of 10 replications for the moistened seed and 4 replications for dry seed.

Five Types of Seed at 4 Water Contents. This experiment was designed to document feeding damage rates to all 5 types of seeds which had been included in earlier experiments, to eliminate differences in feeding rates among ants from different colonies, and to include final seed germination as an index of damage. Natural wheat, corn, sorghum, cotton, and soybean seed were studied sequentially using red imported fire ants from only 1 colony for all replications with 1 type of seed. Seed water contents ranged from dry to 20, 40, and 60% dry basis. As before, seed water contents were monitored by periodically weighing seed from 3 replications. Visible seed damage assessments and germination counts were made at 24, 48, 72, and 96 h. At 96 h, all seeds were placed in petri dishes on wet filter paper to complete germination. Damaged seeds were germinated separately from undamaged seed. Three replications were conducted simultaneously.

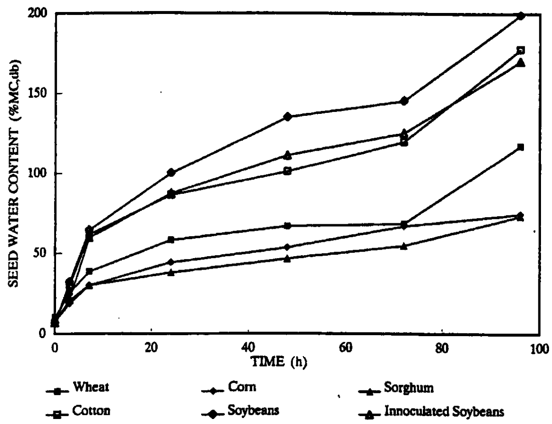

Fig. 1. Water imbibition by seed during the red imported fire ant feeding trial. Dip in curves at 72 h was caused by a procedure anomaly of partial drying before petri dishes were rewetted.

Two arbitrary ratios were calculated for making comparisons as follows: (1) the wet seed/dry seed damage ratio compared the damage rate (percentage per hour) for moistened seed with the damage rate for dry seed (percentage per hour), which has been fairly well documented as reviewed above. A damage ratio of 1.0 would have indicated no change in seed damage between the dry seed and moistened seed conditions; (2) the damage rate/germination rate risk ratio served as an index of the final impact of visually assessed seed damage from feeding, relative to the actual seed germination. The risk ratio was calculated by dividing the seed damage rate (percentage per hour) by the germination rate (percentage per hour) over the same period of time. The main value of the risk ratio was for comparison of relative values between treatments; a higher value indicated a higher impact of visually assessed seed damage on seed germination.

The effects of seed water content on seed damage and seed damage on final germination were examined by analysis of variance (ANOVA) (SAS 1988), with treatment means separated by the Tukey studentized range honestly significant difference (HSD) test, by the Duncan multiple range test, and by regression analysis of trends (Lotus 1992).

Results and Discussion

The addition of water to crop seed served to increase generally the rate of seed damage from red imported fire ant feeding. Imbibition of water seemed to make the seed swell in size and to soften the pericarp, making the seed more susceptible to damage.

Maximum Rate of Seed Germination. Natural soybean seeds tended to absorb water at the fastest rate (Fig. 1) and sorghum seed at the slowest rate. There was some difference between the 2 soybean treatments, with a tendency for slower water imbibition by inoculated soybean. It is interesting that the seeds which produce dicot seedlings (soybeans and cotton) had water contents approximately double those for the monocot seed (Fig. 1). This is discussed later in relation to feeding damage.

Variation in red imported fire ant feeding damage to seed germinating on wet filter paper was highly variable (Table 2). Some tests with wheat and both natural and coated sorghum were 100% damaged within 48–72 h. Conversely, coated corn and all cotton and soybean seeds were damaged <15% during the 96-h test. Not all visually assessed damage inhibited final seed germination. Examination of the statistics (Table 2), revealed that seed damage rate during the 1st 48 h was partitioned into 2 groups of ≈1.0–1.3%/h and 0.10–0.20%/h, with only coated sorghum at an intermediate rate of 0.67%/h. Damage rates for moistened seed were from 3 to 90 times the damage rates for the same seed when dry. The largest increases were for coated cotton and natural soybean seeds, which might indicate an increase in seed coat penetrability or palatability. The damage ratios (wet seed/dry seed) of 2.8–5.5 for all of the other treatments indicates a generally increased ability of red imported fire ants to damage water-softened seed.

Final germination at 96 h approached 100% for the least damaged seed, but was reduced to a range of 65–80% for the highly damaged seed treatments (Table 2). Germination rate over the first 0–48-h period

Table 2. Range of final seed damage for 96-h test, and seed damage, germination rates, and resulting risk ratios for 48-h period when crop seed were under conditions for maximum rate of seed germination

Seed treatment[a]	% range of final 96-h damage	Damage rate for 0–48 h, %/h	Damage rate of dry seed,[b], %/h	Damage ratio of wet:dry seed damage	% final 96-h germination	Germination rate for 0–48 h, %/h	Risk ratio of damage rate/ germination-rate
Wheat (n)	20–100	1.32a	0.47	2.8	78.7ab	0.95b	1.4
Corn (n)	30–80	1.00a	0.27	3.7	99.3a	2.10a	0.5
Corn (c)	2–10	0.10b	0.04	2.5	97.3a	1.82ab	0.1
Sorghum (n)	10–100	1.35a	0.30	4.5	64.7b	0.99b	1.4
Sorghum (c)	4–100	0.67ab	0.14	4.8	80.7ab	1.30ab	0.5
Cotton (n)	4–6	0.11b	0.02	5.5	98.7a	1.98ab	0.1
Cotton (c)	2–8	0.09b	0.001	90	92.0a	1.79ab	0.1
Soybean (n)	6–14	0.20ab	0.005	40	98.7a	1.55ab	0.1
Soybean (c)	4–8	0.11b	0.04	2.8	98.7a	1.76ab	0.1

Means in a column with the same letter are not different at the 5% level of significance in a Tukey studentized range (HSD) test.
[a] n, uncoated seed; c, with a coating as described in Table 1.
[b] From Morrison et al. (1997).

Table 3. Regression coefficients for declining sorghum seed water content with elapsed time

% initial seed water-content group	Replication nos. for regressions	Constant value	Coefficient for time	Coefficient for time²	Goodness of fit of regression, R^2
20	1–5	21.11	−0.271	0.00209	0.89
20	6–10	19.22	−0.361	0.00283	0.85
40	1–5	35.28	−0.726	0.00524	0.94
40	6–10	33.10	−0.888	0.00680	0.92
60	1–5	40.69	−0.858	0.00599	0.98
60	6–10	36.24	−0.941	0.00725	0.95

Table 4. Rates of damage to coated grain sorghum seed 4 initial water contents during the 1st 48 h of exposure to red imported fire ant feeding

% initial seed water-content group, dry basis	Damage rate for 0–48 h,%/h	Damage ratio of wet/dry seed damage
Air-dry	0.04b	NA
20	0.59ab	14.8
40	0.82a	20.5
60	0.88a	22.0

Means with the same letter are not different at the 5% level of significance in a Duncan multiple range test.

was similar for natural and coated treatments of each type of seed. The ideal situation for minimizing the risk of red imported fire ant damage would be to have a seed with low risk ratio (damage rate/germination rate). Coated corn, cotton, and soybean, and natural cotton and soybean did have low risk ratios of 0.1 and would be at less risk than the natural wheat and sorghum seed with comparatively high risk ratios of 1.4. Risk ratio also is useful to compare the effects of commercial coating treatments; the risk ratio was improved from 0.5 to 0.1 for corn and from 1.4 to 0.5 for sorghum by the use of stored product insecticide coatings. There were no risk ratios effects for coatings on the less-damaged cotton and soybean seeds.

Four Levels of Sorghum Seed Water Content. Sorghum seeds moistened to a higher initial water content of 20, 40, or 60% were not statistically different for feeding damage because of large differences among 10 replications (Table 4). Low feeding damage rates for air-dry sorghum seeds, unlike results reported by Morrison et al. (1997), were not explained, but such low rates also occurred for some replications of prewetted seed. As in *Maximum Rate of Seed Germination*, above, feeding damage tended to level after ≈48 h of elapsed time. The damage ratios were 3–4 times higher at 14.8–22.0 than the damage ratio of 4.8 for rapidly germinating coated sorghum seeds (Table 2).

The seeds gradually dried from their initial water contents during the 120-h test because of exposure to ambient air in the vented feeding trays. Such seed drying may occur to shallow-planted seeds in drying soil or where furrows crack open after planting. Rate of drying was characterized by 6 quadratic regression equations, with high R^2 fits of 0.85–0.98 (Table 3).

Table 5. Red imported fire ant 0–48-h feeding damage, final damage, and final germination for 5 types of natural seed; germination is partitioned between damaged and undamaged seed

Natural seed type	% initial seed water-content group	% mean actual seed water content	Damage rate for 0–48 h, %/h	% mean final damage	% final germination of undamaged seed	% final germination of damaged seed	% mean final germination	Damage ratio of wet/dry seed damage	Risk ratio of final damage/final germination
Wheat	Air-dry	10.6	0.15b	12.7b	91.1a	35.0a	84.0a	NA	0.15
	20	20.4	1.17ab	70.7a	97.2a	26.5a	46.7ab	7.8	1.51
	40	42.1	2.06a	99.3a	33.3a	6.1a	6.7b	13.7	14.80
	60	56.3	1.69a	88.7a	29.4a	27.2a	31.3b	11.3	2.83
	—	—	(1.08)	(36.9)	(100.9)	(51.8)	(40.1)	—	—
Corn	Air-dry	8.0	0.63b	64.7ab	100.0a	67.9ab	79.3a	NA	0.82
	20	19.5	1.28ab	73.3ab	100.0a	50.0b	63.3a	2.0	1.16
	40	38.2	0.64b	43.3b	98.6a	65.5ab	84.0a	1.0	0.52
	60	49.1	1.74a	93.3a	66.7a	82.5a	83.3a	2.8	1.12
	—	—	(0.67)	(42.7)	(75.5)	(18.1)	(22.3)	—	—
Sorghum	Air-dry	8.3	0.06b	4.0b	88.2ab	0.0a	84.7a	NA	0.05
	20	22.0	0.19ab	9.3ab	89.0a	5.1a	82.0a	3.2	0.11
	40	38.6	0.79a	38.7a	83.7ab	1.3a	53.3ab	13.2	0.73
	60	47.2	0.75a	36.7a	59.4ab	0.0a	38.0b	12.5	0.97
	—	—	(0.67)	(31.3)	(29.0)	(11.9)	(38.2)	—	—
Cotton	Air-dry	7.0	0.03b	2.7c	69.8a	100.0a	70.7a	NA	0.04
	20	24.3	0.31a	18.0ab	66.6a	28.3b	59.3a	10.3	0.30
	40	47.9	0.44a	25.3a	72.5a	38.0b	64.0a	14.7	0.40
	60	65.1	0.32a	17.3b	87.1a	49.5b	80.7a	10.7	0.21
	—	—	(0.21)	(7.3)	(28.6)	(42.1)	(24.1)	—	—
Soybean	Air-dry	7.1	0.04c	2.7c	98.6a	83.3a	98.0a	NA	0.03
	20	26.1	0.39b	18.7bc	*	*	*	9.8	*
	40	41.7	0.57b	30.0b	61.5b	17.3b	48.0c	14.3	0.63
	60	72.8	0.89a	48.7a	90.1a	68.0a	79.3b	22.3	0.61
	—	—	(0.29)	(17.1)	(17.7)	(48.4)	(18.3)	—	—

Means in a column within each seed type with the same letter are not different at $P = 0.05$, df = 8, ($F_{0.05}$ is given in parenthesis for each group), in a Tukey studentized range (HSD) test. *, Procedure failure; df = 6.

These equations were used to calculate the declining seed water contents for each data time step. A generalized equation to model the sorghum seed damage because of elapsed time of feeding and calculated declining seed water content, is as follows: Damage (%) = −10.4+26.7*log(time, (h))+0.619*(water content (%)), $R^2 = 0.24$.

This model may be useful in defining the general trend of interactions between elapsed feeding time and seed water content under laboratory conditions, even if the regression fit is low at only 0.24.

Five Types of Seed at 4 Water Contents. When natural wheat, corn, sorghum, cotton, and soybean seeds were subjected to procedures as in *4 Levels of Sorghum seed Water Content*, above, red imported fire ant feeding damage generally increased with increased initial seed water content (Table 5) and length of exposure time (not shown). Actual mean seed water contents differed from the targeted values, especially from the 60% highest water content, and may have impacted the specific results but not the trends. Highest final feeding damage was 88.7–99.3% for wheat and corn seeds. In contrast, air-dry cotton and soybean seeds suffered only a 2.7% final damage. The 0- to 48-h damage ratios were generally in the range of 10–15 for seeds with 40% and 60% initial water contents. Exceptions were low damage ratios for corn and higher damage ratios for soybean seeds at high water contents.

Final germination was partitioned between undamaged and damaged seeds. In some cases, damaged seed exhibited 100% germination subsequent to the 96-h feeding trial (Table 5) (e.g., the air-dry cotton). In contrast, damaged sorghum seed, with ≈60% initial water content had a 0% final germination rate. Trends were not consistent, but for the 2 crop seeds most susceptible to red imported fire ant feeding damage (wheat and sorghum) (Morrison et al. 1997), mean final germination decreased with higher initial water contents. Increases in the risk ratio showed that from 2 to 20 times more damage occurs when seeds at elevated water contents are exposed to the feeding of red imported fire ants.

In conclusion, seeds that have adequate available water for maximum rate of germination will suffer some feeding damage, but rapid germination, in combination with some degree of protection (3–5 times reduction of the risk ratio) with tested stored product insecticide–fungicide coatings, may provide adequate germination for economically acceptable crop stands. These data confirm findings of Drees et al. (1991, 1992). If planted seeds gain some increased water content from the soil, but not enough to support germination, the rate of feeding damage and resulting germination will be a function of the seed type, stored product seed coatings, seed water content, and time of exposure to the red imported fire ant. Some more susceptible seed types, including wheat and grain sorghum, may be damaged enough by feeding to sustain complete crop-stand failures. The risk ratio (seed damage rate/seed germination rate) was a useful index for the comparison of the impact of red imported fire ant feeding when comparing differences between species of germinating crop seed, seed water contents, and/or seed coatings.

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