

## CHAPTER 27

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## Nuptial Flight Studies of Field-Collected Colonies of *Solenopsis invicta* Buren

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### Abstract

Procedures are described for field studies of nuptial flights of *Solenopsis invicta* using colonies held captive in soil in 5 gal. buckets. The conditions under which the flights took place were similar to those reported for natural flights, i.e., air temperature 27 to 29° C, high humidity over the colonies, and low wind velocity. Attempts to collect queens on sticky traps of Alsynite or aluminum foil were unsuccessful. A few males were collected, but it is not known if they came from the release colonies. It is suggested that the technique, if modified, could be used on a variety of studies on flight behavior and mating of fire ants.

### Introduction

Nuptial flights of the red imported fire ant, *Solenopsis invicta* Buren, usually occur in the spring and summer from 12:00 pm to 3:00 pm, one or two days after a rainfall. Morrill (1974) reported that in June in North Florida, *S. invicta* colonies averaged about 690 alates per flight and that the number of alates participating in nuptial flights in four generally infested habitats was about 462,000 per hectare. Markin *et al.* (1971) found that males left the nest first and that both sexes readily took flight with a minimum of preflight activity. Based on aerial trapping studies, they reported that the majority of males were captured at a height of about 100 meters, but some males were collected as high as 300 meters. Most females were collected at heights of about 60 to 120 meters. The females generally flew less than one mile before returning to the ground; 95% of the queens descending from the flights were mated.

Rhoades and Davis (1967) found that the relative humidity during nuptial flights was never below 80% and no flights occurred when soil temperature at a depth of 4 inches was below 18° C. All flights occurred when ambient temperatures were between 24 and 32° C. Markin *et al*

(1971) reported flights of *S. invicta* in the winter in Louisiana when air temperature ranged from 20 to 26° C. Lofgren et al. (1975) stated that flights have been noted every month of the year and that no correlation between flight activity and cloudiness or barometric pressure has been found. In most cases wind velocity at time of flights was less than 5 mph.

Markin et al. (1971) suggested that females did not land at random. Vinson and Greenberg (1986) appeared to corroborate this conclusion when they found that plastic swimming pools 8 ft. across and covered with tin foil or water collected more queens from flights than pools covered with soil or dark cloth. They also reported that large numbers of queens can be found after flights in swimming pools and the beds of pick-up trucks.

From this review of published observations it is evident that our knowledge of *S. invicta* nuptial flight activity is scanty and much of it is anecdotal. Because of this we investigated the possibility of "artificially" inducing mating flights in which environmental conditions prior to flight could be controlled. The technique was used in conjunction with a preliminary experiment on site selection by queens alighting from their mating flight. This paper presents the results of these experiments.

## Materials and Methods

### Ant Colonies

*S. invicta* colonies were collected near Gainesville, Florida, March 8-12, 1982 at mid-morning, a time when alates and sex brood were close to the mound surface. Each colony was placed in a five-gallon plastic bucket and transported back to the laboratory. The upper inner sides of the buckets were treated with Fluon® to prevent ant escape. An effort was made to collect colonies with alate males and females; however, one sex usually predominated. A total of 42 colonies were obtained.

The buckets were held in the laboratory at a temperature of  $27^{\circ} \pm 2.5^{\circ}\text{C}$  and a relative humidity of  $60\% \pm 5\%$ . This was done to increase foraging activity and feeding behavior. Following two days of acclimatization, each colony was offered 5.0 gms of pregel defatted corn grits containing 2.5% oil soluble Calco® blue dye dissolved in once-refined soybean oil. This procedure was used to mark the ants internally and enable us to identify any alates captured after their flight. Additional bait was offered to the ants when the previously applied bait had been consumed. Twice a week, a 50:50 honey/water solution

(5 gms) was offered to the colonies. One hundred mls of tapwater were added to the buckets every week to maintain a moist environment. This practice was stopped after three weeks to allow the top few inches of soil to dry. The bottom half remained moist throughout the experiment. After two weeks in the laboratory, the buckets were covered and the colonies moved outside underneath a rain awning until the field releases were made on April 13 and 14. This was done to acclimate them to ambient temperatures. Food continued to be provided as before. Ten days prior to the nuptial flight experiment, 25 alates from each bucket were removed and squashed on gridded-white paper to determine if they had taken the dyed food.

#### Experimental Release Site

The study area for the experiment was the University of Florida's Experimental Beef Research Facility north of Ocala, Florida. Within the site, a 1.94 km<sup>2</sup> area was designated for release and trap location. This area had already been subdivided into nine pastures (ca. 0.64 km<sup>2</sup> each) which facilitated measurements for trap location.

The release site was centered in the study area, 0.96 km from each side. The first designated trap position was placed on the northern boundary and all other trap locations measured from that site. Distance between adjacent trap sites was approximately 201 meters. Trap locations were moved slightly if the originally marked site was obstructed from total sunlight by trees, buildings, or other objects. The trap locations were marked with stakes with a total of 50 locations, including the release site.

#### Trap Design

Two types of sticky traps were used to collect alates descending from nuptial flights. Since *S. invicta* workers exhibit peak light spectrum responses at 360 and 620 mu (Marak and Walken, 1965), it was believed that traps strongly reflecting these wavelengths might attract flying queens. Williams (1973) reported that translucent Alsynite® fiberglass panels were highly attractive to stable flies. Subsequently, Agee and Patterson (1983) revealed that the source of the attraction was the panels' ability to reflect ultraviolet light. Because of these reports, fiberglass was used for one of the trap types. Aluminum foil was used in the second type since it reflects most wavelengths, including infrared radiation.

Four by eight ft. sheets of Alsynite were cut into 50 pieces, (0.71 m<sup>2</sup>). The aluminum foil was cut into slightly larger dimensions and taped,

shiny-side up, to 0.61 m<sup>2</sup> pieces of pegboard. One trap of each type was placed at every trap location. Paint rollers and roller pans (Fig. 1) were used to apply a thin coating of a hexane Tacktrap® solution (6:1 ratio). Previous laboratory tests with different ratios indicated this quantity offered both ease of handling and application. Traps were slightly angled from the horizontal to capture the sun's rays as it crossed the sky.

#### Field Release Procedures

Based on predicted weather conditions favorable for mating flights, the colonies were taken to the experimental site and arranged in decreasing concentric circles, with a maximum outer circle diameter of approximately 4.8 meters (See Fig. 2). The bucket tops were dusted with talcum powder to insure that no ants and alates would crawl out of the buckets. Cockroaches, killed by freezing, were provided as nourishment. Between 4:00 and 4:30 pm, approximately 708 mls of tapwater were slowly and evenly poured over each bucket. This was equivalent to about half an inch of rainfall as calculated by comparing bucket surface area with a Springfield 12.0 cm plastic rain gauge. Twenty wooden tongue depressors were implanted five centimeters in the soil of each bucket to simulate grass and other objects onto which the alates climb prior to flight (Fig. 3,4). Since the interaction of soil moisture and relative humidity required to produce flights were unknown, the soil surface and air space from soil surface to bucket top was lightly misted with approximately 10 mls of tap water. This created a humid microenvironment within the bucket. Rhoades and Davis (1967) reported natural nuptial flights occurred when the relative humidity exceeded 80%.

Air and soil temperature, relative humidity, and ground wind speed and direction were taken at the time of flight. Helium-filled weather balloons were periodically released to ascertain upper wind direction and any upper wind currents.

#### Trap Collection

According to Markin et al. (1971), mated females were found descending to the ground within half an hour after taking flight. This observation was supported by aerial collections, which netted only 58 females and over 3,000 males. Our traps were checked after 24 hours in the event males descended over a longer time period. Fire ant alates found on the traps were placed in medicine cups, brought back to the

laboratory and immediately squashed to determine the presence of dye.

## Results and Discussion

### Nuptial Flights

Weather conditions 24 hours after colony placement in the field on May 13, 1982 appeared to be conducive for mating flights. Overnight temperatures varied between 13 and 16°C, and had warmed to 23°C by 9:30 am. Relative humidity at this time was 61% with light winds from the southeast at 3-5 mph. Average soil temperature 7.6 cms below the soil surface within the buckets was  $20^{\circ} \pm 6.6^{\circ}\text{C}$ . Each bucket was misted between 11:30 and 12:00 noon. At approximately 2:00 pm, preflight behavior (excited workers on surface opening exit holes for alates) was observed in 5 buckets. Air temperature at this time was 27.8°C with relative humidity at 56%. Average ground wind speed had increased to 5-7 mph and its direction was west-southwest. The average soil temperature in the five buckets was  $28.5^{\circ} \pm 3.9^{\circ}\text{C}$ . At 3:15 pm, some female alates from a single bucket took flight. At this time, average wind speed had increased to 7 mph with gusts from 10-15 mph. Weather balloons released at 3:15 pm and 3:30 pm traveled horizontally at about a 20° incline for the first 30 meters, then almost vertically for approximately 91 meters, and finally horizontally to the east at a slight upward angle. Female alates leaving the bucket traveled with the prevailing ground wind current. Flight activity lasted no more than 20 minutes. Alates did not fly from the other buckets exhibiting preflight behavior. Since swarming workers and alates on the tongue blades were above the bucket level, they were exposed to the ground wind current and this presumably influenced the quick cessation of preflight and flight activity. Following this abbreviated flight, no female alates were caught on the sticky traps.

The weather and soil conditions present on the second day are shown in Table 1. As before, all buckets were misted between 11:30 and 12:00 noon. Preflight activity commenced at 12:45 pm in a number of buckets. Actual flights began around 1:15 pm and continued till 3:30 pm. Males were the first to leave the buckets, followed shortly (within half an hour) by females. A total of 32 of the colonies (80%) contributed alates to the mating flight. Of these, 17 contributed males and 15 females. Of the non-contributing buckets, 4 had females only, 3 had males only, and 1 had both males and females. Due to the low (relative to mating flight conditions) prevailing relative humidity, the air directly over the buckets was given a general misting every twenty

minutes from 1:30 to 3:30 pm. This misting resulted in temporary flight cessation which quickly resumed after the mist dissipated.

Average wind speed from noon till 2:20 pm was 5 mph, with some gusts up to 10 mph. These gusts reduced flight activity greatly. About one hour into the mating flights, in an effort to maintain flight activity, a motor van was placed close to the buckets, blocking much of the surface wind around them (Fig. 2). This appeared to increase flight as well as stimulate preflight activity in other buckets. Weather balloons released during this time indicated a moderate wind current at ground level. The balloons rose slowly but steadily until they were out of sight. The alates generally took off into the wind, with most of them flying with the wind upon reaching a height of approximately three meters. The flights began to decrease around 2:45 pm, with only five colonies releasing alates at 3:00 pm. By 3:30 pm, all preflight and flight activity had ceased.

#### Trap Catches

All traps were checked approximately half an hour following the end of the mating flight and again the next day. Markin *et al.* (1971) reported that males often stay aloft until sunset, and may not be collected on the day of the mating flight.

A total of 22 alate males were caught; 64% were trapped on the aluminum foil traps and 72% were collected downwind. No female alates were captured. When returned to the laboratory and crushed on white bond paper, no blue dye was detected from any of the ants. Prior to release, 95% of the alates examined were marked with dye. All of the queens contained large amounts of dye in their crop, however the males were only faintly dyed in their head and thorax. Since males are marked very faintly with the dyed oil procedure, it is possible that our squash technique was inadequate for determining the presence of dye in the males we captured.

The area surrounding our test site was very lightly infested with imported fire ants (<1 mound/hectare). The native fire ant, *Solenopsis geminata* (F) was quite abundant within and around the study area. Although morphological differences between alates of these two species have been described by Hung, *et al.* (1977), it was difficult to observe these characteristics when the specimens are covered with tacktrap. The captured males were totally black, characteristic of *S. invicta*, while *S. geminata* is usually yellowish-brown to dark brown.

It is not known why only male alates were captured in the traps. Vinson and Greenberg (1986) indicate that fire ant queens are attracted more to shiny, reflective surfaces than to soil or dark cloth. In their

study, aluminum foil was distinctly more attractive than other surfaces, with water being second. Their traps consisted of 4.9 meter diameter pools over whose surface the test material was placed. It is possible the traps in our test were too small (61 sq. cm) and/or the number of female alates participating in the flight too small for some of them to view the traps. The traps themselves were inclined only slightly toward the sun, and perhaps did not present an effective reflective surface. It is not known what ratio of females to males existed within the mating swarms.

Although fewer males were captured on Alsynite traps, the sample is not sufficient to support the hypothesis that one reflective surface is more attractive than the other. A smaller number of larger, somewhat elevated traps in a heavy, naturally infested area of *S. invicta*, might provide more conclusive information on flight range and site selection.

### Conclusions

Mating flights were readily induced outdoors from field-collected *S. invicta* colonies held in 5-gal. buckets. The parameters that influenced behavior were similar to those already cited in the literature (air temperature of 27 to 29°C with a high humidity over the release bucket and a low wind velocity). The ability to induce nuptial flights presents unique research opportunities to a) study micro-environmental conditions affecting preflight and flight behavior of *S. invicta*, b) measure the effects of bait chemicals on flight capabilities of reproductives, and c) provide a useful bioassay method for studies of pheromones associated with flight initiation and male/female attraction in flight. The results of the site selection experiment were inconclusive. Only males were captured, predominantly on the high-infrared reflecting surface. None of the males captured were marked with the blue dye incorporated into food given the experimental nests although this may have been caused by the inadequacy of the dyed food technique for marking males. Failure to capture females may be attributed to a) insufficient female alates in mating flight, b) formation of the swarm outside the experimental area, and c) inadequate trap size and orientation.

### Acknowledgements

We acknowledge the superb technical assistance of Dorman Hicks and Ken Plumley. The critical comments of the manuscript made by C. E. Schreck and B. J. Smittle were very helpful and are appreciated.

We thank the personnel of the University of Florida's Experimental Beef Research Station for allowing us to conduct this research on their farm.

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Table 1. Measurements of some environmental parameters made during mating flights on April 14, 1982.

<u>Environmental parameter</u>	Noon	1PM	2PM	3PM
Avg. air temperature (°C)	25.6±0.4	27.2±0.2	28.3±0.3	28.9±0.3
Avg. soil temperature in buckets (7.6 cm) beneath surface	27.1±4.3	30.2±2.9	31.5±2.2	31.1±3.0
Avg. rel. humidity (%)	55.8±1.4	47.6±1.4	45.8±1.6	49.0±2.4
Avg. wind speed (ground, mph)	5.0±6.5	5.0±4.8	4.5±3.9	2.5±0.5
Prevailing wind direction	NW (swirling from ENE)	W	SSW	W (swirling from NW)



Figure 1. Application of Tack-trap-hexane solution to plexiglass trap.



Figure 2. Buckets containing ant colonies in field test. Applicator sticks have been placed in the soil and vehicle has been placed up wind from buckets to shield them from the wind.

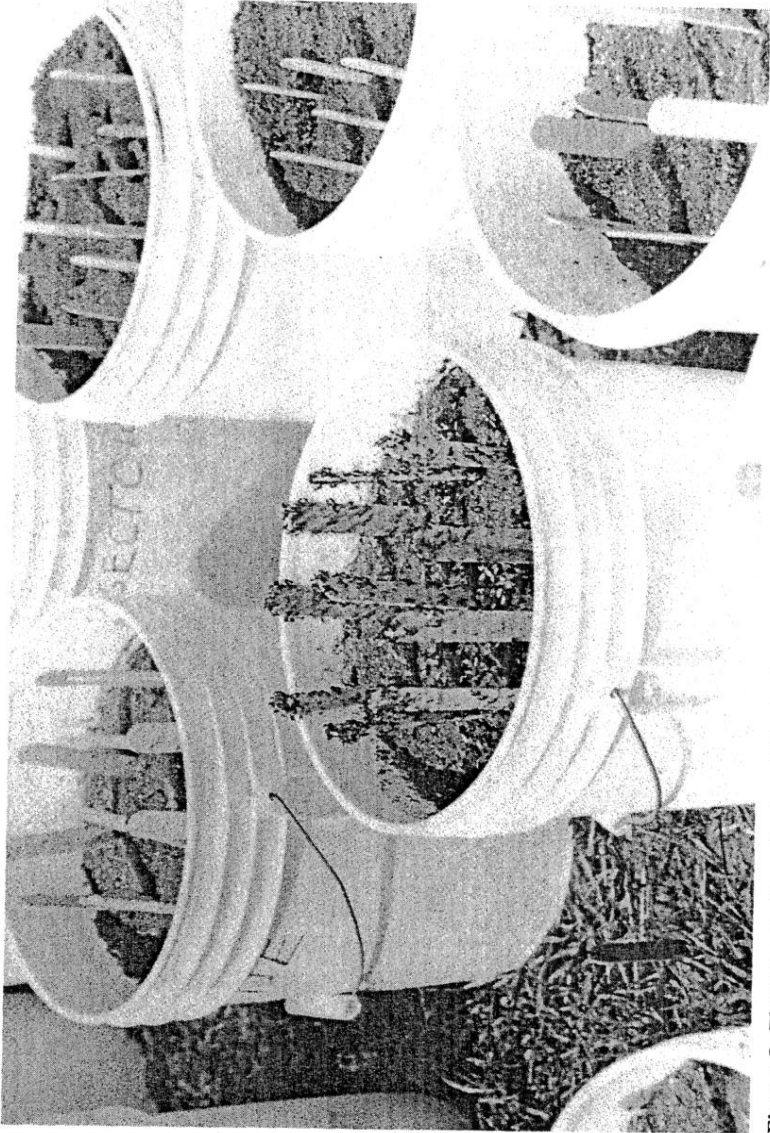


Figure 3. Placement of applicator sticks in the soil showing alates crawling into them.



Figure 4. Close-up view of female alates on applicator sticks prior to flight.