Behavioral Studies of the Southern and Tawny Mole Cricket

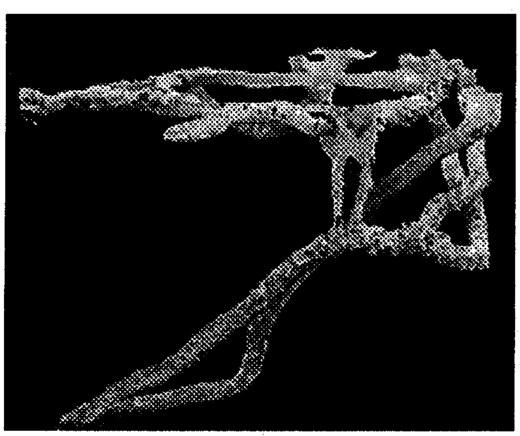
R. L. Brandenburg
Dept. of Entomology
Box 7613
N. Carolina State Univ.
Raleigh, NC 27695
919/515-8876

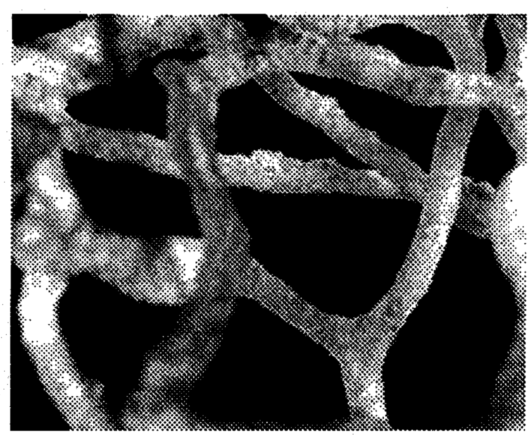
M. G. Villani
Department of Entomology
N. Y. S. Agric. Expt. Sta
Cornell University
Geneva, NY 14456
315/787-2342

Project Description: The goal of this project is to develop a more effective and ecologically sound management strategy for both the tawny and southern mole cricket. The specific objectives of this proposal are to apply this new knowledge by 1) documenting the site preference, dispersal, predation, avoidance behavior, and visible turf damage resulting from intra- and interspecific interactions between individuals in the field and through the use of radiographs, wax castings, and baits in the laboratory and in the field; 2) isolating and characterizing the biologically active compounds that may modify cricket behavior in the laboratory and in the field. The field project at North Carolina State University has allowed us, in concert with the laboratory at Cornell University, to define factors that limit our management capabilities and identify conditions most conducive to effective mole cricket control.

Progress to Date: Radiographic studies of mole cricket tunneling have documented stereotypic behavior of southern and tawny mole crickets. We have determined that soil physical properties, the presence of other crickets (of the same or different species), the presence of biological or chemical insecticides, and the presence of fluids from other crickets can alter this behavior and may help explain the variability observed when attempting to manage crickets in the field.

The use of radiography chambers that are essentially 2 dimensional provides valuable insight into the subterranean activity of mole crickets. However, the actual 3 dimensional components of the mole cricket's behavior are not well documented. The use of larger chambers to hold soil and preparation of a wax-based material to create casts of the tunneling structure has proven quite successful. Was castings provide examples of the use of wax castings to capture the burrowing of mole crickets in large soil arenas. Ordinary canning wax was be heated and poured down cricket tunnels to create permanent wax castings of these tunnels in larger soil arenas. These castings allow us to view and analyze the burrowing behavior of the crickets during their tenure in the soil in response to a variety of control agents under various soil conditions.





These casts document, not only the typical 'Y' shaped structure of the tunnel, but the development of an extensive network of tunnels useful for feeding and escape. They not only confirm radiograph findings, but allow further exploration of cricket behavior. Additionally these arenas are of a sufficient size to determine surface activity and turfgrass damage that is indicative of field damage.

The use of this technique in the field during the summer and fall of 1997 has further documented the accuracy and validity of the laboratory radiographs. Field validation of tawny mole cricket tunneling behavior was conducted by creating wax castings of mole cricket tunnels on a golf course driving ranges which permitted complete excavation of castings. Wax castings in field tunnels and subsequent excavation of these castings have documented the 'Y' shaped tunnels observed in the radiographs. The consistency of these tunnels lends credibility, not only to the laboratory studies, but also to the theory that tunnel construction plays a significant role in mole cricket ecology and avoidance of control strategies.

A Masters student initiated studies on ovipositional preference, dispersal of nymphs and the impact of the southern mole cricket on dispersal on tawny mole cricket and subsequent damage during 1997. These studies have involved intensive sampling of mole crickets (both southern and tawny) over a wide range of soil types, soil textures, soil compaction, soil drainage and moisture characteristics. Data collected include ovipositional preference, nymph abundance and development, and turf damage. These data will help define high risk areas and help target control strategies more effectively. Additional studies on the use of irrigation to modify mole cricket behavior and insecticide activity have been very inconclusive and further justify our research on mole cricket behavior.

Studies on the impact of soil moisture on egg-laying indicate the timing and intensity of oviposition can be significantly affected. This affects not only the likelihood that a mole cricket infestation will occur in a specific location, but also affects the timing of egg laying and ultimate egg hatch. Moreover, this favorable response to higher soil moisture may well serve as a risk indices for high risk areas and thus aid in target monitoring and control strategies. Continued research on mole cricket development as related to soil temperature and acoustic sound trap catches add further support to previously developed population curves.

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The goals of this project were to:
1) improve our understanding of tawny mole cricket and southern mole cricket (southern mole cricket) behavior especially as affected by environmental conditions through radiographic, bait, and destructive studies.
2) isolate and determine the activity of sex, aggregation and alarm pheromones of the tawny mole cricket.
3) determine the behavior of tawny mole crickets in the presence of microbial and chemical insecticides.
4) initiate field studies to better understand tawny and southern mole cricket behavior as suggested by laboratory studies.

Improve our understanding of tawny mole cricket and southern mole cricket behavior especially as affected by environmental conditions through radiographic and destructive studies.

Before detailed studies can be undertaken on the impact of environmental factors and disease status on mole cricket behavior can be undertaken, a clear picture of 'typical' tawny mole cricket and southern mole cricket behavior must be understood. Studies were begun using radiographic technology (x-rays) to visualize the movement and feeding patterns of both tawny mole cricket and southern mole cricket in the soil matrix. Through the placement of a small lead tag on each cricket, tunnel construction and cricket movement in the tunnel could be monitored over extended periods of time.

A series of time lapse radiographs over a twenty-six day period (figure 1: a through p) shows the stereotypical behavior of a single late instar tawny mole cricket nymph. This nymph produces a characteristic 'Y' shaped tunnel that allows two escape routes to the surface and down into the soil to escape predators including larger southern mole crickets, and a long tunnel into the soil profile that most likely aides in thermal and water regulation. Tawny mole crickets typically forage at the root/soil interface between the 'Y' arms and are therefore always near an escape route. As tawny mole crickets grow, their burrows widen and extend further into the soil profile suggesting a possible cause for the difficulty in bringing older crickets to the surface through soap flushes and baits. Crickets also seem to maintain their tunnel system, rebuilding collapsed tunnels over time.

When there is more than one tawny mole cricket in an area each cricket builds its own tunnel system that they maintain over time (as seen in figure 2A[a-d]). There appears to be little burrow sharing between tawny crickets in the same in the same arenas. This would be a good strategy for insects that do not usually attack each other but create burrow system that would reduce interference as they feed at the thatch/soil interface. By comparison, the predatory southern mole cricket appears to create less extensive burrows than do tawny mole cricket. Instead, southern mole cricket burrow predominately at the thatch/soil interface perhaps searching for prey items. Since most plant feeding soil insects including tawny mole crickets are found in this soil zone this is a highly adaptive behavior.

An obvious question to ask is if the borrowing patterns of tawny mole crickets are altered when they are in the soil with hungry southern mole crickets, or if the burrowing patterns of southern mole crickets are altered if they are crowded into the same block of soil. Interestingly, the 'Y' tunnel patterns of the tawny mole cricket does not seem to change in the presence of predatory southern mole crickets (as seen in figure 2B [a-d]). Tawny mole crickets appear 'wall-off' their tunnels when southern mole crickets are present but further studies are needed to confirm this behavior. By comparison, live southern mole crickets will move are far away from each other as possible when placed together in a chamber. Figures 2C [a-d] and 2D [a-d] show two different time lapse series of the tunneling patterns of two southern mole crickets of approximately equal size. This and other similar series suggest, but do not conclusively confirm the presence and activity of a chemically-mediated avoidance behavior in this species.

Studies conducted in this project indicate that prey size is a major determinate in the acceptability of prey for southern mole crickets. In these laboratory studies southern mole crickets were presented tawny mole crickets that were small, of equal size, or larger than themselves. Within one week southern mole crickets had killed and eaten all smaller tawny mole crickets while tawny mole crickets of equal or larger sizes survived. One week later, southern mole crickets preyed upon tawny mole crickets of equal size but did not attack larger tawny mole crickets. Tawny mole crickets that were larger than southern mole crickets caged with them were never attacked by their smaller, hungry cousins. Environmental conditions can alter, but do not destroy the stereotypical movement patterns of tawny mole crickets. For example, a subsoil composed of high density sand does not extinguish the 'Y' shaped burrows of the tawny mole crickets but may cause a bending or the burrow (figure 3A [a]) or a termination of the burrow (figure 3A [b]) at the density boundary.

Previous studies of mole crickets tunneling in the soil with laboratory equipment and the use of radiographs have documented the 'Y' shaped tunneling pattern consistent with t tawny mole cricket. The use of chambers that are essentially 2 dimensional provides valuable insight into the subterranean activity of mole crickets. However, the actual 3 dimensional components of the mole cricket's behavior are not well documented. The use of larger chambers to hold soil and preparation of a wax-based material to create casts of the tunneling structure has proven quite successful. Figure 4 (1-6) and figure 5 (A-B) provide examples of the use of wax castings to capture the burrowing of mole crickets in large soil arenas. Ordinary canning wax was be heated and poured down cricket tunnels to create permanent wax castings of these tunnels in larger soil arenas. Figure 4 reveals the burrow of a single mole cricket over a one week period. Soil was removed at 3 inch intervals from the surface to reveal the burrow as it descended into the soil profile. Figure 5 shows a more intricate burrow created by a southern mole cricket confined to a fairly narrow arena. A 'Y' tunnel is clearly visible in both figures.

These castings allow us to view and analyze the burrowing behavior of the crickets during their tenure in the soil in response to a variety of control agents under various soil conditions. These casts document, not only the typical 'Y' shaped structure of the tunnel, but the development of an extensive network of tunnels useful for feeding and escape. They not only confirm radiograph findings, but allow further exploration of cricket behavior. Additionally these arenas are of a sufficient size to determine surface activity and turfgrass damage that is indicative of field damage. The use of this technique in the field during the summer and fall of 1997 has further documented the accuracy and validity of the laboratory radiographs. Field validation of tawny mole cricket tunneling behavior was conducted by creating wax castings of mole cricket tunnels on a golf course driving ranges which permitted complete excavation of castings. Wax castings in field tunnels and subsequent excavation of these castings have documented the 'Y' shaped tunnels observed in the radiographs (Figure 6 [A-F]). The consistency of these tunnels lends credibility, not only to the laboratory studies, but also to the theory that tunnel construction plays a significant role in mole cricket ecology and avoidance of control strategies.

Isolate and determine the activity of sex, aggregation and alarm pheromones of the tawny and southern mole crickets

Tawny and southern mole crickets were collected in North Carolina and transported to the NYSAES, Geneva, New York for laboratory analysis. It was noted that when disturbed, both mole cricket species discharged a oily, highly odorous substance from their abdomen. Discharges were collected for biological and chemical assays in our laboratory:

a) preliminary chemical analysis: a small discharge sample from each cricket species was prepared for analysis through the use of gas chromatography. Although there appeared to be basic similarities in the two species discharges as indicated by overlapping peaks in parts of the GC detection strip charts, there were also clear differences in the southern mole cricket & tawny mole cricket discharges indicating unique compound constituents in the discharges for these two species. We are currently working in concert with electroantennogram analysis to determine which peaks are bioactive and therefore should be analyzed further (identification & synthesis).

b) Behavioral response of mole cricket nymphs to discharge:

Tawny mole cricket discharge was collected on absorbent cotton and placed in soil arenas along with several tawny mole cricket nymphs. Radiographic analysis shows a clear avoidance of tawny mole cricket to areas near the discharge impregnated cotton further suggesting the biological activity of the discharge. Interestingly, live tawny mole crickets placed in the same chamber do no seem to affect the tunnel patterns of their neighbors suggesting that they do not discharge their compounds around other tawny mole crickets.

1. Biological activity & isolation of sex, alarm and defensive kairmones:

Adult southern and tawny mole crickets were dissected in order to remove anal (males and females) and female protodeal glands. crickets were anesthetized with high does of carbon dioxide gas and then chilled for 15 minutes. Dissections were done under sterile saline solution. When carbon dioxide was administered the cricket usually discharged an oily substance that was collected on filter paper for analysis and bioassays. A pair of anal glands and the hind gut were collected from each insect; in addition, the protodeal gland was collected from all females. 1 glands were soaked in hexane for one hour to extract any bioactive compounds. Gas chromatography and mass spectraphotometry of all samples indicated a range of hydrocarbon compounds. Electroantennograms and electropalpograms gave no differential response among the 13 extracts tested. A double tube bioassay to determine if live crickets showed any behavioral response to the various extracts. There appears to be at least two distinct behaviors exhibited by the crickets. Once they entered a tube, they either moved directly to the end and remained there clawing the end cap, or they exhibited a 'backing' behavior. This behavior consisted of backing up and moving forward, usually in the vicinity of the extract coated filter paper. They also appeared to pause and groom in this region. The strongest initial response was the response of the southern mole cricket male to southern mole cricket female protodeal gland extract but even this response was variable. Additional assays will be conducted to verify or dismiss these preliminary results. Interestingly, when male and female southern mole crickets were place in the tube together, the female often approached the male, after which both the male's and female's behavior was altered, becoming agitated, but the pair had minimal physical contact. The female then flipped on her back and dragged her abdomen over the inner tube, releasing liquid onto the tube and then righting herself. The nature and bioactivity of this extract is being investigated. We see no indication of the presence of a long-range male or female sex or aggregation pheromone in tawny or southern mole cricket adults although we continue to investigate that these compounds do in fact exist.

Determine the behavior of tawny mole crickets in the presence of microbial and chemical insecticides.

Field studies conducted during 1995 by RLB suggested that biological and chemical insecticides may alter the behavior of mole crickets thereby affecting the performance of these agents in the field. Preliminary radiographic assays with one synthetic insecticide suggests that tawny mole crickets can sense and avoid high concentrations of the product in soil thereby reducing overall activity. Radiographic experimental designs where crickets could not escape insecticide suggested a decline in burrow construction and maintenance. Figure 3B [a-c] shows the response of tawny mole cricket nymphs were placed in chambers that contained low density sand in the top third of the chambers and high density sand in the bottom two thirds. Additionally, a mole cricket insecticide was incorporated into the upper half of the low density sand in each chamber. This treatment was one treatment in a larger study testing the interaction of insecticides and soil compaction on cricket behavior. The radiographs in figure 3A can be considered the control treatment for this study. One can see that tawny mole crickets avoid that portion of the chamber that contains the incorporated insecticide by burrowing into the high density sand (3B [a]) or by remaining in the untreated low density sand at the surface (3B [b-c]). One should note that the crickets continued to burrow at the root zone in both sides of the chambers. It should also be noted that this behavior did not occur in every insecticide-treated chamber suggesting that the affect may be transient or be in response to only the parent or one or more breakdown products.

Studies suggest that fungal pathogens placed at the soil surface may also repel crickets and cause them to spend reduced time at the surface thereby increasing the possibility that there may be poor overlap of inundative release of fungal pathogens and crickets in the field. Figure 3C shows the behavior of two tawny mole crickets twelve days after their placement in there chambers. Radiograph 3C [a] shows the typical `Y' shaped burrowing pattern of the tawny mole cricket. By comparison figure 3C [b] shows the tunnels of a tawny mole cricket when high concentrations of the fungal pathogen Beauveria were placed at in the root zone. As you can see this cricket has changed its burrowing system and separated its tunnel from contact with the soil containing the pathogen. Similar results were observed with a different fungal pathogen against Japanese beetle grubs using similar chambers.

To further study this phenomenon, a baiting technique was developed to evaluate the possible deterrent effects of the Beauveria against tawny mole crickets. This technique works well in conjunction with radiographic and other nondestructive monitoring techniques.
Crickets used in this study were placed on uniform feeding regime prior to the initiation of the study. All food was withdrawn for 24 hours prior to test to clear the cricket's digestive systems. An attractive bait (laying mash + feeding stimulant) was prepared following Kepner and Yu 1987. Calico Red Dye N-1700 was incorporated into the bait to allow an indirect measure of bait consumption through dissection and spectrophotometric evaluation (Daum et al. 1969). Beauveria conidia was incorporated into half of the baits (treatment) while the other half of the baits were left fungus-free (control). Crickets were individually placed in test arenas containing soil and turfgrass. A known quantity of either control or fungus-treated bait was placed on the surface of each arena. Crickets were to feed for 8 to 12 hours and removed. Half of the crickets were frozen to measure the quantity of bait (and therefore a comparison of feeding between crickets presented with control and Beauveria baits) fed upon during the previous eight hours. The other half of the crickets were removed and placed individually n new arenas containing only soil and turf to evaluate the impact of the fungus on cricket mortality.
To determine the quantity of bait the crickets feed upon the following protocol was followed:

Crickets were dissected and the entire digestive tract was removed (figure 7a shows the dissected that fed on a bait that did not have dye incorporated into it, while figure 7b shows the red digestive tract of a crickets that feed upon diet into which dye had been added. The digestive systems were macerated with forceps and the dye was extracted with acetone. The concentration of red dye in the resulting solution was determined using a spectrophotometer at 517 nm. This technique allowed us to determine that the presence of Beauveria in baits reduced mole cricket feeding by approximately one half when compared to untreated baits. Most, but not all crickets, in both the control and fungus-treated bait arenas fed on some of the bait in their areas but crickets tended to feed on more of the bait if fungus was not present. Some crickets in both treatments did not feed. These results agree with the radiographic studies that suggest that Beauveria can alter tawny mole cricket behavior. Monitoring of the crickets not dissected showed the beginning of cricket mortality due to Beauveria infection approximately 21 days post treatment. The study was terminated at 21 days post treatment due to high control mortality but only crickets exposed to Beauveria in baits were killed by the fungus.

Initiate field studies to better understand tawny and southern mole cricket behavior as suggested by laboratory studies.

Field studies in North Carolina have provided significant new information on mole cricket development, dispersal, field behavior, interspecies relationships, and the influence of soil environment on damage and control. This labor intensive effort during the past three years have worked in concert with the laboratory studies conducted at Cornell University and provided guidance and insight for these studies as well as serving as field validation of laboratory findings.

Studies have documented coexistence among the two species of mole crickets despite the predatory nature of the southern mole cricket against the tawny. The consistently earlier egg hatch and development of nymphs is a key to survival of the tawny in areas inhabited by southern mole crickets. Southern mole crickets will aggressively feed on tawny mole crickets that are the same size or smaller. The nymph emergence curve for tawny mole crickets is generally two weeks ahead of the same curve for southern mole crickets. However, the nymph emergence curve appears to be truncated due to mortality of late hatching nymphs that are preyed upon by larger southern mole crickets. This overall nymph emergence curve, its timing and duration is critical to effective management of both species of crickets.

Since behavior is influenced by nymph size and since the initiation of control strategies is affected by egg hatch the relationship that has been established soil temperatures and degree day accumulation and the occurrence of these events is important new information. This will help target management strategies to those most susceptible stages as well as providing insight into the best timing to diminish the likelihood of mole cricket behavior minimizing the control strategies effectiveness. In addition, the duration of the egg hatch can be predicted and lead to improved follow-up scouting and management efforts. A manuscript documenting this research and its application is planned within the next year.

Additional research on the effect of soil moisture on egg hatch and surface damage helps us determine when visible surface damage is most likely and when environmental conditions favor significant egg survival in nonirrigated areas. Research data taken under various soil moisture and soil type regimes and in conjunction with and without pesticide use provides us with circumstantial evidence on mole cricket behavior and the response to insecticide use which can be confirmed in the laboratory.

This information has also helped us determine preferred areas of egg laying for both species and is providing significant insight into the identification of "high-risk" areas to help reduce scouting time and develop guidelines for targeting the use of new insecticides which are most effective when used in a preventive mode (e.g. Merit (imidacloprid) and Chipco Choice (fipronil)). These data, in conjunction irrigation studies on the effectiveness of control strategies, is allowing us to document the expected effectiveness of these strategies under a range of field conditions. This is essential information for targeting control efforts to those conditions that encourage maximum effectiveness and minimize overall pesticide use. This is particularly useful for the effective use of biological control materials.

A Masters student initiated studies on ovipositional preference, dispersal of nymphs and the impact of the southern mole cricket on dispersal on tawny mole cricket and subsequent damage during 1997. These studies have involved intensive sampling of mole crickets (both southern and tawny) over a wide range of soil types, soil textures, soil compaction, soil drainage and moisture characteristics. Data collected include ovipositional preference, nymph abundance and development, and turf damage. These data will help define high risk areas and help target control strategies more effectively. Additional studies on the use of irrigation to modify mole cricket behavior and insecticide activity have been very inconclusive and further justify our research on mole cricket behavior.

Studies initiated in Year 3 on the impact of soil moisture on egg-laying continued during Year 4 and indicate soil moisture affects the timing of egg laying and ultimately egg hatch. This favorable response to higher soil moisture is being used to develop risk indices for high risk areas and thus aid in targeting monitoring and control strategies. The second year results from the moisture studies are a component of the new graduate student's research program. This research used 7.5 cm diameter by 15 cm deep PVC oviposition chambers filled with a uniform mixture of oven dried Kureb fine sand. Three soil moisture regimens of 4, 7 sand 10 percent soil moisture were established and individual mated female crickets added to the chambers. The soil was monitored daily for the presence of eggs through day 48. Results demonstrate that higher soil moisture increase the percentage of females that lay eggs as well as the number of eggs per female.

Research on mole cricket development as related to soil temperature and acoustic sound trap catches adds further support to the previously developed population curves from 1994-1996. The relationship between mole cricket development and soil temperature (degree day accumulation) and the use of acoustic sound trap captures of adults female mole crickets and the monitoring of soil moisture is creating a more accurate means for mole cricket forecasting. In addition, studies over the past four years have documented that regardless of the timing of the initiation of egg hatch, the completion of emergence of small nymphs occurs at the same time each year. This consistency of the completion of egg hatch is important from a management perspective.

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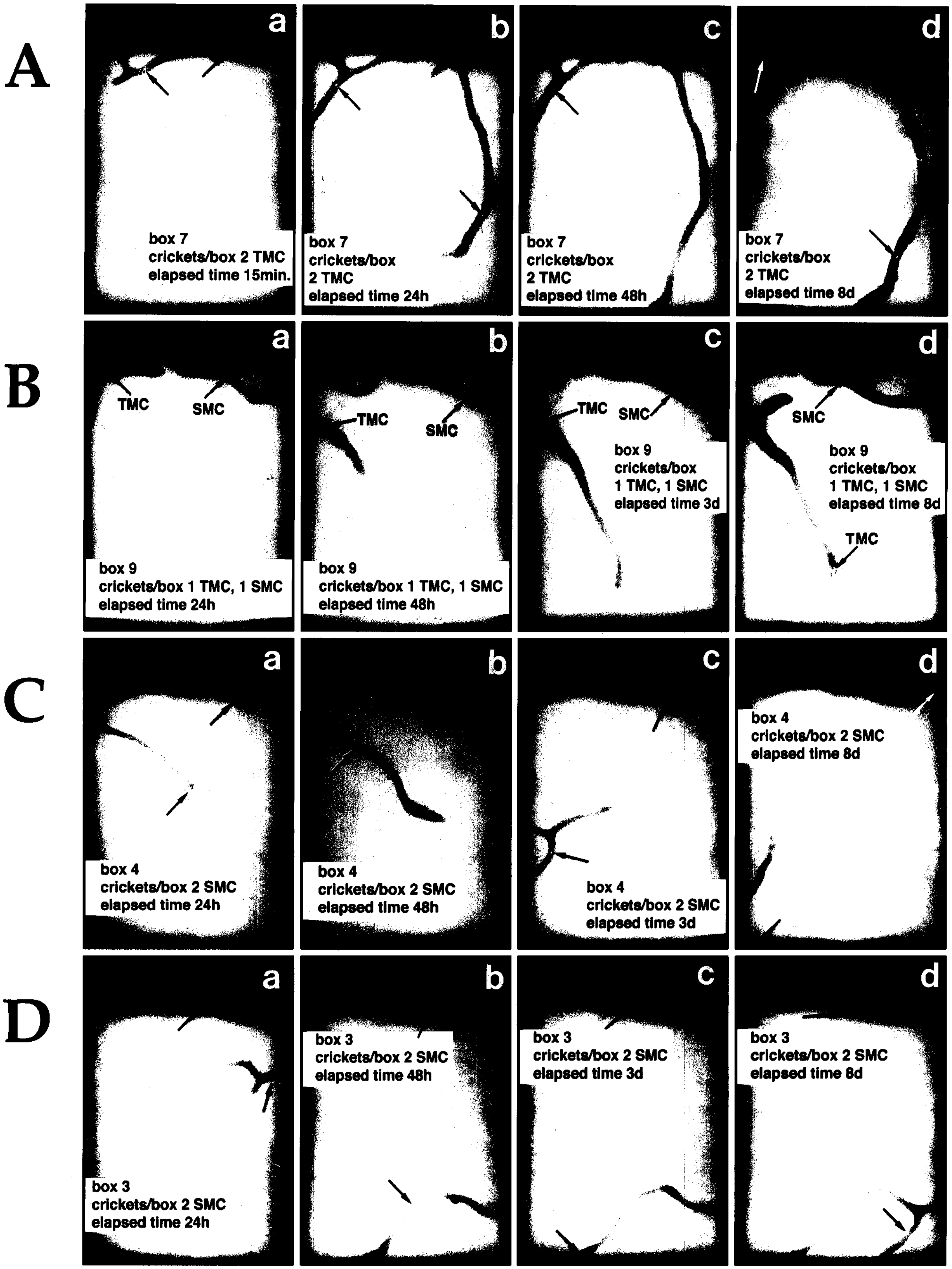

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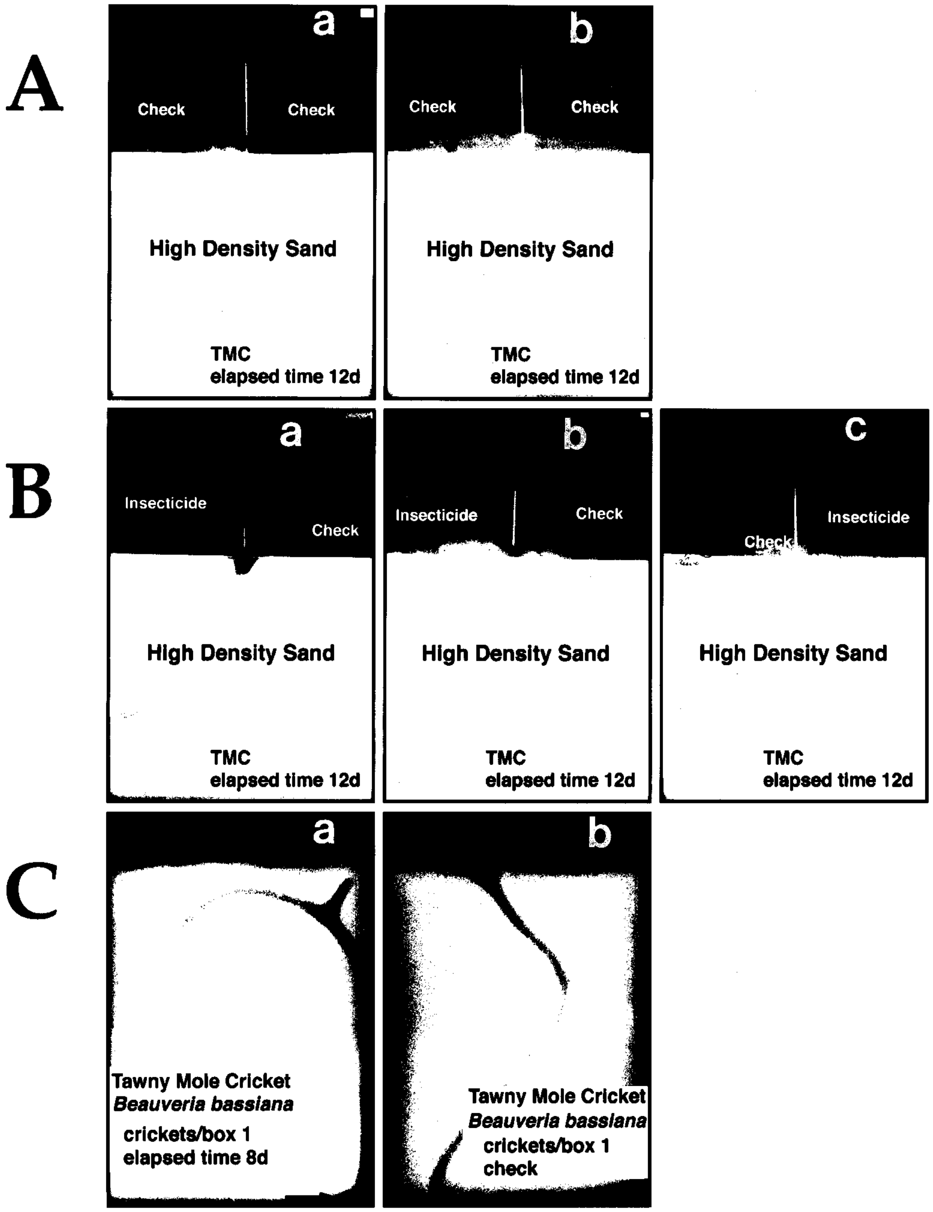
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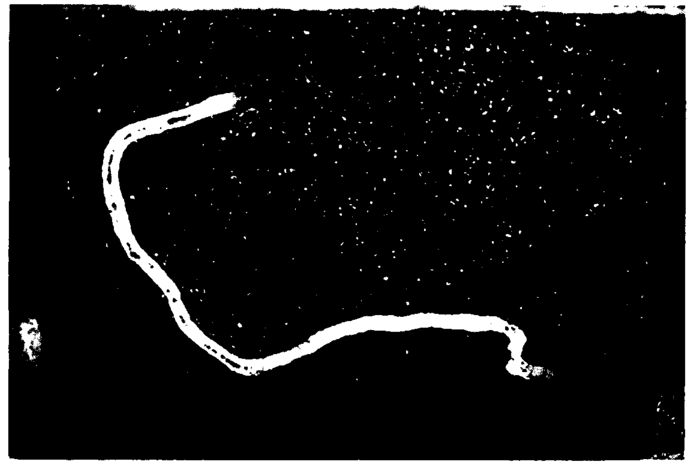

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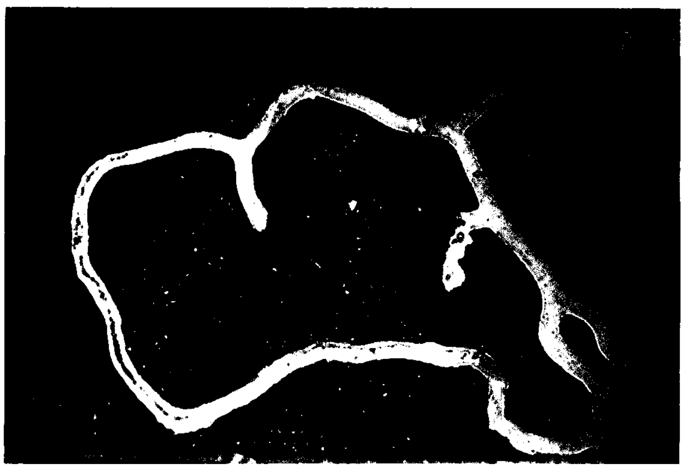

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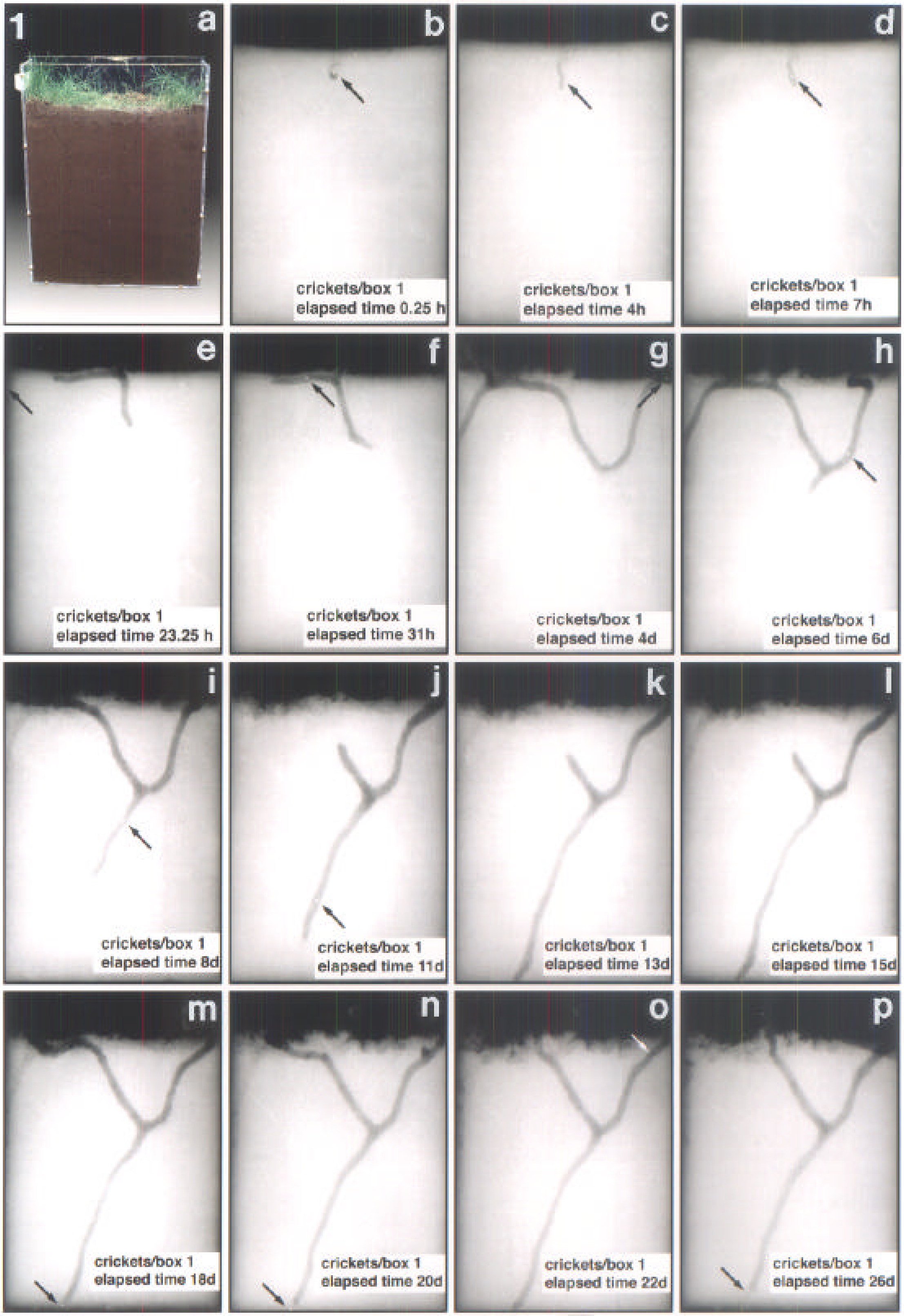
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