PROPOSAL FOR FY2003-2004 COMPETITIVE GRANTS PROGRAM

Texas Imported Fire Ant Research and Management Project

16 Apr 2003

Title of project: Identification and Development of Grasses to Repel or Significantly Reduce Red Imported Fire Ant Density in Pastures, Conservation Lands, Urban Lawns, and Recreational Areas.

Lead Principal Investigators

James A. Reinert, Ph.D., Professor of Urban Entomology, Texas A&M University Res. & Ext. Center, 17360 Coit Rd. Dallas. TX 75252- 6595; Ph: 972-231-5362; E-mail: j-reinert@tamu.edu

Gad Perry, Ph.D., Assistant Professor, Dep. of Range, Wildlife, & Fisheries Management, MS 2125, Texas Tech University, Lubbock, TX 79409-2125; Ph: 806-742-2842; E-mail: gad.perry@ttu.edu

Co-Principal investigators

Carlton Britton, Ph.D., Professor, Dep. of Range, Wildlife, & Fisheries Management, MS 2125, Texas Tech University, Lubbock, TX 79409-2125; Ph: 806-742-2842; E-mail: carlton.britton@ttu.edu

Ken Helms, Ph.D., Research Scientist, Dep. of Entomology, Texas A&M University, College Station, TX 77843. Phone: 979-862-8645; E-mail: khemls@tamu.edu

Paul Paré, Ph.D., Assistant Professor, Dept. of Chemistry and Biochemistry, Texas Tech University, Lubbock, TX 79409-1061; Ph: 806-742-3062; E-mail: paul.pare @ttu.edu

S. Bradleigh Vinson, Ph.D., Professor of Entomology, Dep. of Entomology, Texas A&M University, College Station, TX 77843. Phone: 979-845-9754; E-mail: bvinson@neo.tamu.edu

Summary

Red Imported fire ant (RIFA) populations are often largest in grass-dominated systems such as pastures, conservation lands and in urban lawns, parks, golf courses and other recreational areas, yet can differ greatly between different grassland sites. Our research has found that these differences can be caused by the occurrence of different grass species and cultivars, as well as by differences in watering regime and plant height and density. The reasons for these differences are diverse. Our preliminary research suggest they include both direct effects on the ants (differences is food provided, direct repellency, and prevention of colony establishment) and indirect effects, via the insect prey that provides them with proteins and sugars. The research we are proposing will focus on the relative importance of these factors. It will combine a correlational approach, in which existing sites throughout the RIFA's range in Texas will be evaluated, with controlled small-scale studies in which parameters will be carefully controlled. In addition, analyses of plant chemistry will determine the bases for RIFA repellency or attraction and should have applications to chemical management of RIFA. By identifying species of grass and management practices that result in significantly reduced RIFA densities, this study will provide new biological approaches for ranchers, homeowners, and managers of golf courses, parks and school grounds for effective long-term reduction of their RIFA populations.

Background information

The economic impact of introduced organisms in the U.S. alone has recently been estimated at over $137 billion each year (Pimentel *et al.*, 2000). One of the most noxious exotic pests has been the Red Imported Fire Ant (RIFA, *Solenopsis invicta* Buren; Vinson, 1997). Pimentel *et al.* (2000) estimated the annual damage caused to the U.S. economy by this species to be approximately $1 billion per year. In Texas annual financial costs of damages and expenditures attributed to RIFA was $1.2 billion based upon a statewide survey by Lard (2001). Damage depends on the density at which RIFA occur, and population sizes can differ greatly within, as well as between, locations. Unfortunately, RIFA densities are often greatest in pastures, conservation lands and in urban lawns, parks, golf courses and school grounds. Damage to Texas agriculture is estimated at $90.6 million (Lard, 2001), and the cattle industry is impacted at an estimated $255 million per year (Teal *et al.*, 1999). The greatest loss attributed to RIFA in Texas was in urban areas, with an estimated $702 million in cost to residential households, followed by cemeteries ($63.9 million), golf courses ($45.9 million), institutions ($47.3 million), schools ($42.3 million) and commercial businesses ($45.9 million) (Lard, 2001). The RIFA continues to spread in Texas, and these numbers are likely to increase, although there are some indications that ecosystems may be able to partially accommodate to RIFA over time (Morrison, 2002). No methods for large-scale eradication of RIFA are currently available. As a result, development of methods of biological control is one of the highest priorities of the Texas Imported Fire Ant Research and Management Plan (available at http://fireants.tamu.edu/plan/). An array of such methods currently is in development, focusing on diseases, parasites, and predators of RIFA (L.E. Gilbert, pers. com.; D. F. Williams, pers. com.; H. Thorvilson, pers. com.) We are proposing a somewhat different approach, which will result in identification of species and cultivars of forage and turf grasses repellent or deleterious to RIFA populations, as well as management practices, that can result in significantly reduced RIFA densities. For example, in a recent experimental investigation of the causes of microgeographic differences in RIFA densities, we have discovered highly significant differences among 20 different grasses replicated in plots located in a Linear Gradient Irrigated System (Reinert *et al.*, manuscript in prep.). In this study, differences in landscape management practices (including irrigation and mowing practices and species and cultivars of turf grass) were highly significant. RIFA colony establishment increased as the soil moisture decreases to ca. 60 to 70% replacement with irrigation. Additionally, for selected turf cultivars, there were often 9 to 12 colonies established in the half of the plot maintained at 2 in. but only 1 to 3 colonies in the half cut at 1/2 in. Major differences were also identified among the four turf species of *Buchloe Cynodon, Stenotaphrum, and Zoysia* as well as among the cultivars. Understanding why populations are small at particular locations and in particular grasses and landscapes, while larger at others, will provide new insight into how managers can reduce RIFA populations in various landscapes and environments.

Fire ants are omnivorous, feeding on almost any plant or animal material, although insects and other small invertebrates appear to be their preferred food (Vinson & Greenberg, 1986) and are essential for normal larval growth (Sorensen *et al.*, 1983; Williams *et al.*, 1987; Porter, 1989). RIFA colonies require large amounts of sugars, which appear to be a key element determining population size (Porter, 1989; Williams *et al.*, 1980; Helms and Vinson, 2002; Hallway *et al.*, 2002). These sugars come primarily from plant sap, either directly (by ants chewing on plants or from extrafloral nectaries), or indirectly (from honeydew exuded by plant sucking Homoptera: aphids, scale insects, and mealybugs). Sugars contained in these liquids are probably essential in fueling workers activities because workers are incapable of ingesting solid food (Glancey *et al.*, 1981). Recent research suggests that, because of the role that different plants play in RIFA nutrition, differences in RIFA density may be related to differences in plant species. Fire ants attack young saplings and seedlings, destroy buds and developing fruits, and have been shown to feed on the seeds of 139 species of native wildflowers and grasses (Lockley, 1997). Damage to plants is exacerbated during periods of drought, when fire ants seek alternate water sources. Because grasses are the major plants in pastures, conservation areas, lawns, golf courses, and other recreational areas, they are the likely source of sugars fueling RIFA populations.

A study conducted in 2001 shows that pastures where bermudagrass occurs have much larger RIFA populations than adjacent fields where bermudagrass is absent (Helms and Vinson, manuscript in prep.) A separate study (Britton et al., 2003) evaluated RIFA populations in and around pastures planted with Old-World bluestem grass WW-B.Dahl (*Bothriochloa bladhii*), a high-yielding, warm-season, perennial bunchgrass used in Texas for premium hay and grazing (Sanderson *et al.*, 1999). Incidental observations suggest it is less likely than other grasses to be infested with RIFA. As a preliminary test of this observation, we surveyed a limited number of pastures and determined the population density of ants in several types of forage (Britton et al., 2003). We measured 30.5 and 30.8 RIFA colonies per acre on shortgrass prairie and mixed shortgrass-bermudagrass pastures on the High Plains respectively. On an adjacent field sowed in 10-ft., alternating rows with B.Dahl and another Old World Bluestem over a decade ago, we measured only 0.05 colonies per acre. A pure B.Dahl field across the road had no visible RIFA colonies. In Lampasas County there were 33.0 RIFA mounds per acre in a bermudagrass field. Two adjacent B.Dahl fields, surrounded with areas of very high RIFA density, had 12.9 and 0.0 colonies per acre. These limited data suggest that fields of B.Dahl are indeed infested with far fewer RIFA colonies. The usefulness of B.Dahl hinges on two main issues. First is *biological efficacy*, which will be determined by the present study. Although the effect of this grass species on RIFA populations has not been previously studied, some preliminary work does support the pattern we have observed, whereby RIFA are excluded or reduced in B.Dahl fields. Several authors have noted that some members of the genus *Bothriochloa* are less attractive to insects than others (Zalkow et al., 1980), suggesting some species "are resistant to the impacts of some insect pests" (Pinder and Kerr, 1980). These and other authors studied the volatile oils that give these grasses their distinctive smell and identified a range of oil-based components. Whereas some landscaping materials are known to have limited efficacy in repelling RIFA (Thorvilson and Rudd, 2001; Anderson *et al.*, 2002), the potential impact of B.Dahl remains unstudied. The ability of the isolated chemicals produced by B.Dahl to repel insects also has not been tested.

A second crucial issue is *cost effectiveness*. Luckily, WW-B.Dahl is one of the most inexpensive improved grasses to seed. Although the per-pound seed costs are high (2003 estimate is about $16 to $18/ lb. PLS), the seeding rate is only 1 lb. PLS/ac (compared to, for example, about $50/lb for native seed mixes). Similar to other species of grass, a clean, firm, seedbed is needed to enhance B.Dahl seeding success. Grazed-out wheat pasture is a perfect seedbed for B.Dahl as long as the area is not disked. With proper management, both B.Dahl and bermudagrass fields should not need reestablishment for a long period of time, probably 30+ years. Comparing seeding B.Dahl to sprigging 'World Feeder' bermudagrass, another improved grass with similar forage production potential, clearly illustrates how inexpensive B.Dahl is. Using a single 100 acre field for comparison and assuming the seedbed preparation costs are the same, B.Dahl will cost about $1,800 for seed and $1,000 for seeding (at $10/ acre). On the other hand, World Feeder bermuda sprigging at 25 bushels per acre will cost $25,000 and sprigging will cost $3,500 for the same field (www.worldfeeder.com/Sales/html). Thus, the cost for seeding B.Dahl is $28.00/acre, compared to $285/acre for World Feeder Bermuda. In addition, Bermudagrass fields may have to be treated with an herbicide several times during the establishment phase. In contrast, weed control is not as important for B.Dahl, a strong competitor once it is established. In addition, maintenance expenses of bermudagrass field will be comparatively high. Following establishment, WW-B.Dahl will require about 100 pounds of N/acre in Central Texas to achieve maximum yield, while World Feeder bermuda will require about 4 times that amount for a similar yield (C. Britton, unpublished). Finally, if the biological efficacy of B.Dahl is established, additional savings will be provided in avoiding the expense of both RIFA treatment and reduction of direct damages to wildlife and livestock caused by RIFA.

The utilization or development of RIFA-resistant grasses is one approach for management of this pest. As grass species are identified with RIFA resistance, additional genotypes can be evaluated from cooperating grass breeding programs. These include: bermudagrass (Taliaferro at OSU, Stillwater, OK and Hanna, UGA, Tifton, GA); buffalograss (Riordan at UNE, Lincoln, NE); Paspalums (Burson at USDA at College Station, TX, Burton Tifton, GA, and Duncan at UGA, Griffin, GA); zoysiagrass and bentgrass (Engelke at TAMU-Dallas); and bluegrass (Read at TAMU-Dallas). Seed sources of additional grass species; *Panicum* spp.; *Sorghastrum sp.*; *Festuca sp.*; *Bothriochloa sp.*; and *Bouteloua sp.* can also be obtained. We propose to evaluate and develop alternative grasses that may significantly reduce the RIFA in grassland habitats (pastures, lawns, and golf courses) and to leverage funds within this funding period to demonstrate that we can exploit those differences to reduce the impact of the RIFA.

Hypotheses, Objectives, and Proposed Work

Global hypothesis 1: Certain grasses can be identified that are antagonistic, repellant or resistant to the establishment of the red imported fire ant.

Global hypothesis 2: Certain management practices associated with grasses can be identified that reduce the establishment and/or prevalence of the red imported fire ant.

Objectives: To test the two principal hypotheses above, we have identified four specific objectives which will be addressed in the proposed study. In general, work on turf grasses will be spear-headed by TAMU personnel and pasture grass work will be led by TTU scientists.

Objective 1: Experimentally determine the degree to which different species and cultivars of grasses used for pasture and turf affect RIFA population size and colony densities. (Reinert and Helms)

The correlational study described below will provide a broad-based analysis of actual RIFA densities under relevant field conditions. To substantiate grass preferences, we will use an experimental setup in a closely-controlled greenhouse experiment. We (Dr Helms) will examine the effect of various turfgrasses (bahiagrass, bermudagrass, blue grama, bluegrass, buffalograss, centipedegrass, tall fescue, zoysiagrass, and St. Augustinegrass) and pasture grasses (Old World bluestems, bermudagrass, dallasgrass, Kleingrass, swithchgrass, indiangrass, sideoats grama, eastern gamma, Texas bluegrass, tall fescue with and without endophyte, little bluestem, big bluestem and native grass mixes) on the growth of captive RIFA colonies. However, we will focus on the turf grasses this first year, as resource cuts will require a reduction in effort. We will select candidate cultivars and genotypes for potential food to include different levels of known insect resistance. We will raise small RIFA colonies from foundress queens collected following mating flights. We will then allow each RIFA colony to forage on a potted grass. Grasses will be grown in greenhouse containers, and each colony will have access to only one pot. During the experiment, nutritional carbohydrates will be collected by the ants from the selected plants. However, we will feed each colony similar amounts of insect prey in order to supply proteins.

Biomass of the RIFA colonies will be determined prior to the start of our experiment. After approximately three months, all colonies will be collected, and any gain or loss in colony biomass will be determined. Statistical analyses of changes in colony biomass will indicate whether and to what degree the different plant species and varieties contributed to colony growth. We are currently cultivating many of the turf and pasture grasses in the greenhouse at TAMU. We are also able to easily rear the necessary RIFA colonies, and we anticipate no problems with conducting this experiment. (We hope to have some of the turf grasses evaluated within the next year).

Multiple-choice studies will be conducted primarily by Dr Reinert. In these studies we will allow IFA`s to forage into multiple grass selections in the laboratory to identify the ants preferred or non-preferred grass species and cultivars. Each replicate will include at least six selected grasses in a randomized arrangement. After multiple selections, several grasses should be identified that are either non-preferred or repellent to RIFA foraging.

Objective 2: Test whether there are differences in RIFA mound densities in Texas pastures and lawns planted with different grass species. (This objective will be a focus of the Texas Tec. group)

The test will involve a large-scale correlational study involving multiple RIFA infested areas of Texas. At each site we will investigate at least two adjacent areas to generate paired comparisons among the densities of RIFA mounds in pastures and lawns planted with various grass species. In each case we will record grass species and RIFA mound density. To increase statistical power, we propose to survey at least 20 sites across the fire-ant infested part of the state, sampling at least five plots and at least two grass species at each site. RIFA population densities will be estimated in native pastures, bermudagrass pastures and B.Dahl by baited cup survey (Morrison, 2002) and by the circle transect method where each mound is also rated (Lofgren and White, 1982). The survey of RIFA abundance will be analyzed by repeated measures ANOVA. An ANOVA design with plant type as the independent variable and RIFA mound density as the response variable will be used to analyze our findings, with a critical *P* value set *at 0.05*. Because of the large size of the state and the infested area, and the need to adequately cover a variety of environments (see below), a relatively large portion of the requested budget for TTU (below), which will lead this part of the study, is devoted to travel expenses.

Given the promising preliminary data involving B.Dahl, we will place a special emphasis in our survey on this pasture grass. Consequently, more detailed data will be utilized to compare the biology of RIFA in B.Dahl and non-B.Dahl plots. For example, we will use the methods of Harlan et al. (1981) and Lofgren and Williams (1982) to assess the level of RIFA activity inside and outside of mound. These data will be analyzed using a repeated measure design and will provide insight into the mechanism underlying differences in RIFA abundance.

Objective 3: Determine if the RIFA resistance observed in select grasses (turf and forage) is the result of direct insect repellency or a reduction in available sugar and protein resources. (these studies will begin later in the first year to allow us to focus on identifying first the more resistant and susceptible grasses).

The two studies described above will identify plant species which reduce RIFA mound density and activity in the field, or colony growth under controlled conditions. In the series of studies that follows, our aim is to identify the mechanisms for this effect.

In *Experiment 1* (this will be started at the end of the first year if continued funds are anticipated), we will provide small RIFA colonies a choice of nesting in either of two 12 X 12 in. trays planted with either a resistant or a susceptible turf or forage cultivar or genotypes (based on our replicated plot results) devoid of insects to determine if there is any direct repellency. If so, we will determine whether repellent cultivars are effective as RIFA repellant mulches by determining repellency using leaf and stem material layered over the soil. This work will be carried out using captive colonies at TAMU.

Whether directly (i.e., by attracting or repelling them) or indirectly (e.g., by keeping out RIFA and decreasing its impacts), the grasses identified as bioactive might have effects on additional organisms as well. In *Experiment 2*, we plan to remove RIFA colonies from replicated plots at TAMU where colonies differed significantly in density according to grass species/cultivar, grass height, and watering regimes (Reinert et al. manuscript in prep.). However, we will put off this project to the second year. If funds are provided the second year we will then use pitfall traps and visual surveys to collect and monitor the insects that might serve as food for the RIFA in resistant and susceptible cultivars and species of either turf or forage grasses. This will be done on a monthly basis, and will complement our research on the importance of different grasses in providing plant carbohydrates by determining whether different grasses differ in the degree they supply the RIFA with proteins in the form of insect prey.

Our *Experiment 3* uses a correlational approach to address the same issue under field conditions. (This will be initiated by the Texas Tec. group, but the effort will be reduced by more of a focus on fewer sites.). We will choose a subset of the B.Dahl sites for more intensive study and use standard sampling techniques (e.g., Thompson *et al.*, 1998; Agosti *et al.*, 2000) to assess the biodiversity at each pair of sites. Sampling will be extensive and focus on groups of organisms likely to show a response. For example, we will actively search for horned lizards (genus *Phrynosoma*) and their food organisms, which are known to be adversely impacted by the presence of RIFA (Donaldson *et al.*, 1994), as well as mealybugs known to be associated with RIFA (Helms and Vinson, 2002). At least six paired sites will be evaluated in this manner, and paired comparisons will be used to statistically test the null hypothesis that no difference exists for biodiversity on B.Dahl and other pastures.

Finally, *Experiment 4* will focus on isolating and identifying repellent compounds using bioassay-tracking methods, as well as determining relevant resources such as the sugar and protein content of the grasses. This project will be initiated late in the year as relevant grasses are identified). Grasses that show non-preference or repellency to RIFA in other experiments will be assayed for volatile as well as non-volatile compounds using bioassay driven standard methods in the lab of Dr. Vinson. Researchers at TTU will study chemicals in pasture grasses using a two-stage experimental design. First, a Y-maze behavioral assay will be used to establish whether volatile chemicals are responsible for the RIFA-repelling activity of B.Dahl. By allowing RIFA to choose between air streams containing plant odors released from insect resistant and non-insect resistant grasses, we will initially identify if the repellent properties of particular grasses is due to volatile and/or non volatile plant components. Second, the specific chemicals involved will be isolated if necessary. Plant volatile organic compounds (VOCs) will be characterized in the laboratory of Dr. Paré (TTU) and Dr. Vinson (TAMU) from head-space VOC collections of different grass species and analyzed by GC and GC-MS analysis. Comparisons will be made between VOC profiles for biologically active and inactive grasses to narrow down the potential set of compounds with repellant properties. Non-volatile components will be examined by comparison HPLC profiles of organic extracts from biologically active and inactive grasses. This part of the study could lead to identification of RIFA-repellent mulches and chemicals that can be used in large-scale management efforts.

Objective 4: Determine the impact of environmental factors and turf management practices (fertility, irrigation, and mechanical cultural practices) on the establishment of RIFA on turf and forage grass species and cultivars. (This objective will be the focus of Drs James A. Reinert and James C. Read addressing the points in the first paragraph for this year of funding).

The three objectives above detail studies to address our first hypothesis. Our last objective directly addresses our second hypothesis. Preliminary results have demonstrated that soil moisture and mowing height-of-cut show a highly significant difference in RIFA establishment in various turfgrass species and cultivars. Because commercial turfgrass and residential lawns can be more closely managed, this objective is focused more on urban landscapes. Further, since we have found that high moisture and very short grass is less favorable for the RIFA, we want to determine if these conditions are more important following RIFA mating flights, which can be predicted, or must they be applied over longer periods or throughout the growing season. The Linear Gradient Irrigation System at the TAMU-Dallas Center will be used to further these studies with four grass species (bermudagrass, buffalograss, St. Augustinegrass, zoysiagrass) including 20 cultivars.

In conjunction with the planned large-scale correlational study described in # 1 (above), we will conduct surveys to identify factors, such as soil type, soil moisture, precipitation, and temperature, which might help determine the magnitude of this effect. Data will be obtained from direct measurements in the field (e.g., soil type), as well as the literature (e.g., climatic data). We will use a multiple regression approach, with mound density as the dependent variable and dummy variables representing noncontinuous variables such as soil type. To ensure the comprehensiveness and predictive power of this analysis, we will conduct our surveys over much of the area of Texas currently infested by RIFA. These data will complement preliminary work conducted previously by one of us (Vinson, unpublished data).

Expected deliverables

If B.Dahl or other grass cultivars are indeed shown to have RIFA-suppressing qualities, this will provide a powerful new approach to RIFA management. Planting of such grasses in pastures and other areas is likely to be of special value to the cattle industry. Likewise, the identification of turfgrass cultivars or landscape setting that is antagonistic or repellent to RIFA can be used in lawns, golf courses and similar applications to help alleviate the millions of dollars spent annually for suppression of this pest. At the end of this project, we expect to be in a position to confidently recommend which grass species or cultivars of forage and turfgrass can used to significantly reduce the density of RIFAs in a variety of pasture and landscape applications, and how to best manage these grasses to induce a long-term reduction in RIFA populations. This approach is complementary to others currently under development, and the objectives listed above are all designed to address the issue of efficacy and safety of this potential tool. Specific deliverables will be as follows:

1) Evaluation of the efficacy of multiple turf and pasture grasses in reducing RIFA abundance under both experimental and field conditions (objective #1, 2).
2) Identification of the mechanisms by which this repellency is achieved (objective #3).
3) Detection of additional benefits from use of B.Dahl or other grasses, such as repelling other pests or encouraging survival of species of concern such as the horned lizard (objective #3).
4) Recognition of environmental conditions, such as soil moisture and management practices, which further enhance this repelling effect (objective #4).

These specific deliverables will allow us to develop detailed recommendations on how to best manage grasses for the long-term reduction of RIFA densities under conditions relevant to Texas agricultural producers and land managers.

Timeline (for work to be done and anticipated technology transfer / implementation)

Most of the work described above will be initiated as soon as funds are received. This includes recruitment of two graduate students (one at TAMU Dallas/College Station, the other in the department of RWFM at TTU); identification of appropriate study sites; beginning of field sampling; and initiation of most controlled studies. Most of the work will be completed during the second project year, with final reports and technology transfer generally expected towards the end of the second year. A more detailed timeline for each of the objectives is given below.

	Months 1-3	**Months 4-6**	**Months 7-9**	**Months 10-12**
Objective 1	**Initiate**	**Continue**	**Continue**	Complete
Objective 2	**Initiate**	**Continue**	**Continue**	Complete
Objective 3			**Initiate**	**Continue**
Objective 4				**Initiate if additional funds are anticipated**

Relevance to the Texas Imported Fire Ant Research and Management Plan

The overall goal of the *plan* is "to develop products and procedures to reduce RIFA populations to a level that eliminates this insect as a serious pest in terms of economic losses and health threats." Specifically, the *plan* calls for development of "new techniques" and "integrated pest management solutions". The approach we advocate is aimed at delivery of an effective, ecologically sound tool for control or suppression of RIFA and is thus directly relevant to these goals. Our research will do this by identifying which grass species and management regimes can be used in ranching, lawns, parks, school grounds, golf courses or other recreational or conservation area can achieve a significant reduction in RIFA densities. Further, we will explore the mechanisms by which the reduction in RIFA

References cited

Agosti, D., J. D. Majer, L. E. Alanso, and T. R. Schultz (Eds.) 2000. Ants: Standard Methods for Measuring and Monitoring Biodiversity. Smithsonian Institute Press, Washington DC.

Anderson, J.T., H. Thorvilson, and S.A. Russell. 2002. Landscape materials as repellents of red imported fire ants. Southwestern Entomol. 27:155-163.

Britton, C., G. Perry, C. Villalobos, R. Kellison, C. Craig, H. Thorvilson, V. Allen, and B. Racher. 2003. Red Imported Fire Ants and WW-B.Dahl: preliminary observations. Research highlights, Range, Wildlife and Fisheries Management, 2002, vol. 32, p. 22. Texas Tech University College of Agricultural Sciences and Natural Resources, Lubbock, Texas.

Donaldson, W., A. H. Price, and J. Morse. 1994. The current status and future prospects of the Texas horned lizard (*Phrynosoma cornutum*) in Texas. Texas J. Sci. 46:97-113.

Glancey, B. M., R. K. Vander Meer, A. Glover, C. S. Lofgren & S. B. Vinson. 1981. Filtration of microparticles from liquids ingested by the imported fire ant, *Solenopsis invicta* Buren. Insectes Sociaux 28: 395-401.

Harlan, D. P., W. A. Banks, H. L. Collins, and C. E. Stringer. 1981. Large area tests of AC217300 bait for control of imported fire ants in Alabama, Louisiana and Texas. Southwestern Entomol. 6:150-157.

Helms, K.R., and S.B. Vinson. 2002. Widespread association of the invasive ant *Solenopsis invicta* with an invasive mealybug. Ecology 83: 2425-2438.

Holway, D.A., L. Lach, A.V. Suarez, N.D. Tsutsui and T.J. Case. 2002. The causes and consequences of ant invasions. Annual Review of Ecology and Systematics 33:181-233.

Lard, C. F., V. Salin, D. B. Willis, S. Robison, and K. Schroeder. 2001. The statewide economic impact of the red imported fire ant in Texas. A part of the Texas fire ant initiative 1999-2001. Texas A&M University, College Station, TX. Fire Ant Econ. Res. Rep. 01-08: 76 p.

Lockley, T.C. 1997. Imported Fire Ants. World Wide Web URL http://www.ent.agri.umn.edu/academics/classes/ ipm/chapters/lockley.htm

Lofgren, C. S., and C. T. Adams. 1982. Economic aspects of the imported fire ant in the United States, p. 124-128. *In* The Biology of Social Insects, Int. Union for the Study of Social Insects.

Lofgren, C. S., and D. F. Williams. 1982. Avermectin B1: a highly potential inhibitor of reproduction by queens of the red imported fire ant (Hymenoptera: Formicidae). J. Econ. Entomol. 75: 798-803.

Morrison, L.W. 2002. Long-term impacts of an arthropod-community invasion by the imported fire ant, *Solenopsis invicta*. Ecology 83:2337-2345.

Pimentel, D., L. Lach, R. Zuniga, and D. Morrison. 2000. Environmental and economic costs associated with non-indigenous species in the United States. BioScience 50:53-65.

Pinder, A.R., and S.K. Kerr. 1980. The volatile essential oils of five *Bothriochloa* species. Phytochemistry 19:1871-1873.

Porter, S. D. 1989. Effects of diet on the growth of laboratory fire ant colonies (Hymenoptera: Formicidae). J. KS Entomol. Soc. 62: 288-291.

Sanderson, M.A., P. Voigt, and R. M. Jones. 1999. Yield and quality of warm-season grasses in central Texas. J. Range Manage. 52: 145-150.

Sorensen, A. A., T. M Busch & S. B. Vinson. 1983. Factors affecting brood cannibalism in laboratory colonies of the imported fire ant, *Solenopsis invicta* Buren (Hymenoptera: Formicidae. J. KS Entomol. Soc. 65: 140-150.

Teal, S., E. Segarra, C. Barr, and B. Drees. 1999. The cost of red imported fire ant infestation: the case of the Texas cattle industry. Texas J. Agr. Natural Resources 12:88-97.

Thompson, W.L., G.C. White, and C. Gowan (eds). 1998. Monitoring vertebrate populations. Academic Press, San Diego. 365 pp.

Thorvilson, H., and B. Rudd. 2001. Are landscaping mulches repellent to red imported fire ants? Southwestern Entomol. 26:195-203.

Vinson, S.B. 1997. Invasion of the red imported fire ant (Hymenoptera: Formicidae): spread, biology, and impact. Am. Entomol. 43:23-39.

density is achieved, and hope to identify specific compounds which can be used for large-scale RIFA control. Preliminary evidence suggests these approaches are likely to provide a truly new and highly economically beneficial concept of RIFA management.

The *plan* identifies agricultural impacts to be one of the most important problems associated with RIFA. B.Dahl is most commonly used as forage in pastures. Thus, it offers greatest hope for ranchers who already use introduced grasses to graze their livestock, and is supported by the Texas Cattlemen's Association (see attached letter of support, Appendix A). This aspect of the study fits well with an ongoing project on the sustainability of livestock forage systems headed by Dr. Vivien Allen, Thornton Distinguished Faculty of forages at Texas Tech University. The sustainable project already has an established forage grazing system that includes B.Dahl. Dr. Allen has agreed to support this study by allowing us to study the effects of B.Dahl within her forage grazing system (see attached letter of support, Appendix B). In addition, the Texas National Guard has expressed great interest in B.Dahl as a potential mechanism for controlling RIFA on their training sites, especially firing ranges. The Guard is allowing us to use their sites for some of the work described in this proposal (see support letter, Appendix C).

Ecological and recreational impacts are also specifically targeted in the *plan*. The proposed study directly addresses this target by examining the effects of a number of turfgrasses on RIFA abundance. The broad expertise we are bringing to bear on this issue will allow us to determine the effects of grasses on diverse invertebrate and vertebrate populations in a variety of management contexts, including golf courses, schools, and lawns. As a result, the proposed research hopefully will lead to reduced densities of RIFA in multiple environments, resulting in healthier, less stressed domestic animals and reduced RIFA problems in recreation and conservation areas.

Anticipated research results will be published in appropriate State, Regional and National publications. In addition, in order to speed dissemination of findings, results will be presented at the annual Fire Ant Conference and either posters or oral presentations will be presented at multiple other scientific association meetings. Papers will also be presented at various State Commodity Organization meetings.

In summary, the potential long-term benefits from this research will be more efficient suppression of RIFA in agricultural, recreational, state and conservation contexts. It promises to do so while allowing a reduction in the use of pesticides. Thus, the proposed study directly addresses a number of the goals of the *plan*, offering considerable benefits to the citizens of Texas.

Vinson, S. B. & L. Greenberg. 1986. The biology, physiology and ecology of imported fire ants, 193-226. *In* S.B. Vinson. (ed.). Economic impact and control of social insects. Praeger, NY 422 p.

Williams, D. F., C. S. Lofgren & A. Lemire. 1980. A simple diet for rearing laboratory colonies of the red imported fire ant. J. Econ. Entomol. 73: 176-177.

Williams, D. F., K. R. Vander Meer & C. S. Lofgren. 1987. Diet-induced nonmelanized cuticle in workers of the imported fire ant, *Solenopsis invicta* Buren. Arch. Insect Biochem. Physiol. 4: 251-259.

Zalkow, L.H., J.T. Baxter, R.J. McClure, Jr., and M.M. Gordon. 1980. A phytochemical investigation of *Bothriochloa intermedia*. J. Natural Products 43:598-608.

Budget

Texas Tech University:

Perry, Gad and Britton, Carlton:

A. Personnel		
(-)	Principal Investigators (Summer salary)	10,413
(-)	Research Associates	0
(-)	Technician	0
(1)	Graduate Student	19,850
(-)	Student assistants	0
(-)	Laboratory technician	0
B. Capital Equipment		0
Field data collection devices with analysis software		0
C. Travel		
	Domestic. Travel to establish field plots and to harvest data; travel to meetings to present results.	14,100
D. Other Direct Costs		
	Materials, Supplies, field plot prep.	750
	Publications Costs	0
	Miscellaneous supplies	1,250

Budget Justification: *A. Personnel.* Funding is requested to support a variety of functions. Three of the TTU investigators will each devote one summer month to work on this project, primarily in the field. Two graduate students will be recruited, one at each university, to carry out much of the ongoing work. Additional assistance will be provided by research assistants, undergraduate students, and one half-time phytochemistry technician, required to collect, extract, and analyze chemical samples collected from the repellent and non-repellent grasses. The sums listed include base pay and all fringe benefits required by university and departmental rules.

B. Travel. The proposed work contains considerable field-work. Because of the need to provide comprehensive information in a large state that has multiple environments and land uses, extensive travel will be required to assure coverage of representative sites.

Paré, Paul:

[redacted]			
A. Personnel			
	(1)	Principal Investigator	0
	(-)	Research Associates	0
	(-)	Technician	0
	(-)	Graduate Students	0
	(-)	Undergraduate Students	0
	(1)	Ant Maintenance Tec.	0
B. Capital Equipment			0
C. Travel			
	Domestic		0
	Foreign		0
D. Other Direct Costs			
	Materials and Supplies		5,032
[redacted]			

Budget Justification:

Dr Paré will work with Dr Vinson in the isolation and identification of IFA repellents from the repellent grasses.

<u>Texas Agricultural Experiment Station</u>

Reinert, J. A. & J.C. Read:

A. Personnel	
(-) Principal Investigator	0
(1) Research Associate	26,991
(-) Technician	0
(-) Graduate Students	0
(-) Undergraduate Students	0
(-) Secretary	0
B. Capital Equipment	0
C. Travel	0
D. Other Direct Costs	
Materials and Supplies	1,000

Budget Justification

We will be focused on part of Objective 1and 4. Most is for salaries to do the work.

Vinson, S. B.:

[redacted]

A. Personnel		
(1)	Principal Investigator	0
(-)	Research Associates	0
(-)	Technician	0
(-)	Graduate Students	0
(-)	Undergraduate Students	0
(-)	Ant Maintenance Tec.	0
B. Capital Equipment		0
C. Travel		
	Domestic	0
	Foreign	0
D. Other Direct Costs		
	Materials and Supplies	5,149
	Publications Costs	0
	Chemical Analyses	0
	Services	0

[redacted]

Budget Justification

Dr. Vinson. No direct cost to the project. Will direct and oversee the bioassays, obtaining ant material and provide ant expertise. He will work closely with Dr. Reinert (TAMU-Dallas) and Perry and his group (Texas Tec.) to insure coordination with elements of this project with both the Pheromone and the IFA Reproduction projects. The Chemistry part of this project will be conducted by Dr. Williams, who is part of the Pheromone Proposal. Since all the equipment and expertise is part of that program, he has consented to work with us on this project with Dr. Helms conducting the bioassays and helping with the chemical analysis.

Supplies are needed to maintain ants for bioassays, Helms, K. R.:

A. Personnel		
(-)	Principal Investigator	0
(1/2)	Research Associate	15,465
(-)	Technician	0
(-)	Graduate Students	0
(-)	Undergraduate Students	0
(1)	Ant Maintenance Tec.	0
B. Capital Equipment		0
C. Travel		
	Domestic	0
	Foreign	0
D. Other Direct Costs		
	Materials and Supplies	0
	Publications Costs	0
	Chemical Analyses	0
	Services	0

Budget Justification

Dr. Helms will devote half time to the project. He, along with Mrs. Ellison, will conduct the bioassays. He will also work with Dr. Vinson, Dr. Williams (Texas A&M) and Dr. Pare (Texas Tec.) in conducting some bioassays.

<u>SUMMARY BUDGET</u>

Gad Perry and Texas Tec. program	51,395
Reinert	27,991
Vinson	5,149
Helms	15,465
TOTAL	100,000