



Henry—seed survival

NEW PROPOSAL

Title:

Assessment of microbial and environmental factors on jointed goatgrass seed survival in soils from Oklahoma and the Northern Plains

Personnel:

Principle investigators W. Brien Henry¹, Francisco C. Calderon², and Maysoon M. Mikha³. Cooperators: Lynn Fandrich⁴, Phil Westra⁵, Alan Helm⁶ and Merle F. Vigil⁷
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Time Line:

Initiation date: July 1, 2006, Termination date: June 30, 2008.

Hypothesis:

Jointed goatgrass germination is affected by soil temperature and moisture regime and is further impacted by soil microbial influences which may be location and site specific.

Summary of Proposal:

Microbial and environmental influences on JGG germination will be examined in the field and under controlled growth chamber conditions. JGG seeds will be placed on the soil surface and in soils (growth chamber = with or without microbes; field = with and without seed treatment) from Oklahoma and the Northern Plains under a variety of environmental conditions (at optimal moisture and at different incubation temperatures). Following incubation, germination will be measured. If significant differences exist with respect to presence or absence of microbes, additional microbial analyses will be conducted on these soils.

- Objectives:**
- 1) **Determine if soil microorganisms influence germinability of JGG seed.**
 - 2) **Determine if soil temperature affects microbial degradation of jointed goatgrass seed in soil.**

Justification:

Previous research has introduced the concept of “weed suppressive soils” that can be potentially deleterious to weed seedlings and weed seeds (Kennedy et al. 1996). It is possible that management can be adjusted to favor this type of microorganism. Studies have also documented soil microorganisms attacking the seed coat of the weeds (primarily velvetleaf); thus, reducing seed longevity (Kremer 1993). Donald and Zimdahl (1987) examined JGG seed longevity with respect to 5, 10 and 30 cm burial depths and determined that longevity declined rapidly regardless of burial depth. By the fifth year of this study, four of their initial five sites had no viable JGG seed remaining in

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the top 5 cm of soil. Alternatively, Sagar and Mortimer (1976) suggest that wild oat seed viability suffers a significant reduction from 60% if buried an inch or two beneath the surface down to 15% if left on the soil surface between the months of November through March. This research would suggest that burying seeds extends their life; however, differences in jointed goatgrass persistence have been observed in soils exposed to vastly different tillage regimes. In Oklahoma, yearly sub-soiling tillage operations appear to have significantly reduced JGG populations. In Colorado, deep tillage was a solution to jointed goatgrass in the past; however, in order to conserve moisture and enhance cropping intensity, the increase of no-till or limited tillage over the past 10-15 years appears to have favored JGG populations. If microbial degradation of JGG seed varies with respect to soil type or environmental conditions, perhaps we could then use this information to manage soil conditions (tillage) or fertility inputs (C/N ratios) so that the crop is favored over the JGG.

Methods:

Growth Chamber Component: JGG spikelets will be collected from naturally occurring field populations near Akron, CO in August 2006. Spikelets will be kept in a -20 C freezer for a predetermined length of time until initiation of the growth chamber incubation studies. Prior to the complete study, a preliminary study will be conducted on the spikelets to determine dormancy and germination percentage. To address dormancy, a subset of spikelets will be dissected; seed productivity will be documented and seed viability will be assessed via tetrazolium assay. Soils will be gathered from two locations: Oklahoma and the northern portion of the Central Great Plains. Physical properties of the soil such as pH, bulk density, and water holding capacity will be determined. It will be noted from what horizons these soils originate. The management histories, with respect to cropping and herbicide application, will also be noted. Half of these soils will be treated to kill all soil organisms. Microwaving or gamma irradiation are both highly effective in sterilizing soils without damaging soil structure. JGG spikelets will be added to soils with and without microbial activity and exposed to various temperatures in growth chambers or temperature controlled incubators. Following incubation for predetermined period of time, the seed germinated at 25/15 C and 12-hr photoperiod (Fandrich 2005). Germination will be conducted in 4” x 4” clear acrylic germination boxes with a minimum of 14 days for germination. Percent germination will be recorded and if there are significant differences with respect to presence absence of soil organisms, microbial population analysis will be conducted.

Microbial population analysis: At the end of the growth chamber incubations, we will pick seeds that failed to germinate from the non-sterile soils. A total of 50 seeds will be plated on Nutrient Agar amended with cycloheximide, and Potato Dextrose Broth amended with streptomycin. This way, we will attempt to isolate fungi or bacteria involved in the decomposition of JGG seeds. Care will be taken to note the location of microbes (if possible), lemma, palea, or embryo. The plates will then be incubated at room temperature and the bacteria and fungi will be subcultured until pure cultures are obtained. The bacterial and fungal isolates will be identified using the MIDI Sherlock® Microbial Identification System, which identifies isolates based on gas chromatographic (GC) analysis of fatty acid methyl esters (GC-FAME). Note that the Calderón laboratory is equipped to carry out all the microbiological culturing. The microbial identification will be carried out at the Acosta-Martinez laboratory, USDA-ARS.

Field Component: Field studies will be conducted with the same soils. JGG seed will be placed in mesh bags, and buried (50 spikelets per bag) at various depths, including some on the soil surface within PVC pipes. A small metal screen across the top of the PVC pipe will prohibit predation from small mammals. Moisture and temperature will be recorded with watchdog programmable sensors at periodic intervals for each seed burial depth. A commercial seed treatment like Captan or Benlate will be used on half of the treatments to eliminate microbial influence on seed decomposition. Seed will be removed at various time intervals following initiation of the experiment and germinated under ideal conditions in the greenhouse. Following the germination test, the spikelets will be dissected while looking for the presence of seed. If the seed appear healthy, a tetrazolium assay will be conducted. Percent germination will be recorded and if there are significant differences with respect to seed treatment, microbial population analysis will be conducted as mentioned previously.

Statistical Analyses: Data will be analyzed using a split-plot design with temperature as the whole plot factor, and soil moisture, the presence or absence of microbes, and soil type (location) in a complete factorial design within a temperature regime. An analysis of variance (ANOVA) F-test will be used to test treatment factor main effects and interactions. An F-protected t-test will be used to test pair-wise comparisons to follow up any significant findings.

Literature Cited:

- Fandrich, L. 2005. Vernalization and dormancy requirements of jointed goatgrass (*Aegilops cylindrica*). Ph.D. Corvallis, OR: Oregon State University. 164 p.
- Kennedy, A. C., and R. J. Kremer. 1996. Microorganisms in weed control strategies. *J. Prod. Agric.* 9:480-485.
- Kremer, R. J. 1993. Management of weed seed banks with microorganisms. *Ecol. Applic.* 3:42-52.
- Sagar, G. R., and A. M. Mortimer. 1976. An approach to the study of the population dynamics of plants with special reference to weeds. *Ann. Appl. Biol.* 1:1-47.
- Donald, W. W., and R. L. Zimdahl. 1987. Persistence, germinability, and distribution of jointed goatgrass (*Aegilops cylindrica*) seed in soil. *Weed Sci.* 35:149-154.

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Planned Technology Transfer:

Information will be included in presentations at the USDA –ARS Akron Research Station and CCTA (Colorado Conservation Tillage Association). Data will also be published in peer reviewed journals such as Weed Science or Weed Technology and in conservation fact sheets. ~~tillage fact sheets.~~

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BUDGET:

We are requesting \$25,000 total for this two-year project.

ITEMIZED BUDGET JUSTIFICATION STATEMENTS:

BUDGET JUSTIFICATION:

A. Salary and Wages. \$19,200

\$19,200 (\$4,800 times 2) (two-year period) Hourly labor to assist with seed collection, plot maintenance and data collection. 960 hr x (GS-2) ~10.00/hr

B. Fringe Benefits. \$300

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\$300 (two-year period) Summer students.

Benefits rate for hourly student labor calculated at rate of 1.52%

Nonexpendable Equipment. \$0

E. Materials and Supplies. \$4,116 = \$900 + (\$470.19 x 2) + 2,000 + \$276.00

Item	cost
Potato Dextrose Agar	\$85.55
Cycloheximide	\$69.84
Streptomycin	\$40.89
Nutrient Agar	\$110.49
Petri plates	\$163.42
Total:	\$470.19
<u>Total Budget Soil Analysis</u>	
<u>Spring 2006 (One Time Expense)</u>	
\$96.00	Soil Moisture
\$60.00	EC + PH
\$120.00	Organic Matter
\$276.00	

PVC pipe, \$100; germination cubes, \$650; potting soil \$100; collection bags, pens, and other expendable supplies, \$250.

The microbial identification will cost \$100 per sample for 20 isolates, for a total of \$2,000.

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F. Travel. \$284

Trip to gather soil in Oklahoma.

Lodging 1 night at \$75 = \$75

Per diem 2 days at \$50 = \$100

Gas: 600 miles/Dodge 2500 (12 MPG) 50 G of Gas at 2.25 = 112.5\$

G. Publication Costs. \$1,100

800\$ / Article (1 Article), \$800; Fact sheets \$300