

Avra Valley Buffelgrass Seedbank: A Preliminary Report

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Introduction

The City of Tucson (COT) currently owns large parcels of retired cropland in Avra Valley, Arizona. Much of this land has experienced little or no native vegetation recovery, with annual exotics like *Salsola kali* (Russian thistle) often being the dominant vegetation. These lands are the source of numerous ecological and economic problems, including a lack of aesthetic or wildlife value, and often pose a liability issue for the landowner in terms of blowing dust and weeds. Active revegetation of these lands has been complicated by limited economic incentives, low and variable precipitation, extreme temperatures and evapotranspiration, few available propagules, presence of exotic invasive plants, altered hydrology and soil structure, and low soil fertility (WRCC 2000, Roundy et al. 2001).

A past solution to establishing permanent vegetation on the retired cropland in Avra Valley was to seed aggressive African grasses like *Pennisetum ciliare* (buffelgrass), a regulated and restricted noxious weed in Arizona, and one of the few species to establish readily from seed in this environment (Thacker and Cox 1992). The resulting *P. ciliare* infestation has spread rapidly onto surrounding lands, preventing native vegetation recovery and threatening nearby conservation efforts such as the COT Habitat Conservation Plan and preserves such as Saguaro National Park and Ironwood Forest National Monument. *P. ciliare* is widely recognized as a serious threat to life, property, biodiversity, and local economies in the Sonoran Desert. Areas

dominated by buffelgrass support lower abundance and variety of grassland birds and fewer insects than native grasslands (Flanders et al. 2006). Buffelgrass also decreases richness and diversity of shrub and herbaceous plant species (Clarke et al. 2005; Jackson 2005). A full discussion of the issues presented by *P. ciliare* can be found in the Southern Arizona Buffelgrass Strategic Plan, available for download at http://desertmuseum.org/invaders/invaders_plan.htm.

COT has undertaken to control the *P. ciliare* infestation, and is now in the second year of herbicide treatments. Following the successful removal of *P. ciliare*, COT's intention is to pursue active revegetation to establish a permanent cover of native plants on the site (Harold Maxwell, pers. comm.). However, without first eradicating or significantly reducing the buffelgrass seedbank, any revegetation efforts will likely be compromised by the aggressive *P. ciliare* due to its ability to outcompete native plants for water and nutrients. Reduction in the *P. ciliare* seedbank following the mortality of the existing *P. ciliare* plants increased success of native plant establishment in restoration attempts in *P. ciliare*-infested areas in Texas and Hawaii (Tjelmeland *et al.* 2008, Daehler and Gorgen 2005).

This study seeks to support COT's control and restoration efforts by evaluating the trends in *P. ciliare* and native seedbank densities over time as control efforts progress. Specifically, we seek to answer 2 important questions: 1) how long will viable *P. ciliare* seeds remain in the soil and 2) will *P. ciliare* seed in the seedbank be replaced by native seed as *P. ciliare* seed inputs are (presumably) reduced? Trends in the *P. ciliare* seedbank over time should give an indication of the effectiveness of the control efforts and possibly help estimate the length of time such efforts will have to continue to sufficiently reduce the *P. ciliare* seedbank. This will also help verify the assumption that *P. ciliare* seed inputs will directly affect the density of the *P. ciliare* seedbank.

Monitoring trends in the native seedbank will provide important information about the need for active revegetation efforts once the *P. ciliare* is removed.

Our hope is that reduced density of *P. ciliare* plants on COT Avra Valley properties will cause the seedbank of *P. ciliare* to become less dense and less viability over time. Similarly, because we assume that *P. ciliare* densities are sufficient to limit establishment and/or persistence of some native species, and that these species may begin to recolonize following reduction in density of live *P. ciliare* plants, we expect that native seedbank density and viability will increase over time.

Because COT was unable to begin herbicide control in 2006, the sampling design had to be modified to fit the actual implemented control schedule. To answer the questions posed above, baseline (pre-herbicide application) samples were needed for comparison with samples collected post-herbicide application. We collected baseline samples in June 2006 but re-collected in July 2007 after no herbicide application occurred and also because of the above-average summer precipitation in 2006, which could have produced seed that would confound our original baseline samples. In short, at this time we have 2 sets of baseline samples and can therefore infer nothing about the effects of the herbicide control effort. The comparison between these baseline samples is the subject of this report. To answer the research questions and assist the control and restoration efforts on these lands, we strongly urge COT to continue with the original intent of the study and allow for post-herbicide sampling in this and future years.

Methods

At the COT property in Avra Valley near Manville & Reservation roads (Figure 1), Chris Hannum, Ben Wilder, and Travis Bean sampled the seed bank in untreated buffelgrass stands in

June 2006 and three separate areas in July 2007 that had been mowed, sprayed with herbicide, or left untreated. In June 2006 they collected 10 1-dm² samples from each of four buffelgrass plots located randomly within 0.5 mi east and west of Reservation road 1-2 mi north of Mile Wide Rd. In July 2007, 2 pre-herbicidal control treatments (mowing and burning) were performed on the buffelgrass and the sampling strategy was modified to accommodate this. In 2007, 15 1-dm² samples were collected in the mowed and burned areas, and another in an area that was left untreated. A timeline of treatments is provided in Table 1. The sampling device, a metal frame 10 cm × 10 cm × 2 cm, was pounded into the ground until flush with the soil surface. All soil inside the frame was removed to a depth of 2 cm with a trowel and placed in a labeled plastic bag. The bagged samples were stored at room temperature.

Between Feb 2007 and Feb 2008, Chris Hannum, Ben Wilder, and Lynda Klasky processed the samples by sieving them to remove rocks and debris. Chris Hannum and Ben Wilder then used an iterative frothing, flotation, and filtering technique to separate seeds and organic matter from soil particles (Pake and Venable 1996). The organic fraction was then air dried at room temperature and stored in labeled plastic bags. Between May 2007 and Feb 2008, Chris Hannum examined each organic fraction under a dissecting microscope to identify seeds and determine their viability. Shriveled, punctured, broken, cracked, or split seeds were considered nonviable and discarded. The remaining seeds were identified by comparison with voucher specimens. Some seeds of certain genera, notably *Lepidium*, *Euphorbia*, *Mentzelia*, *Plantago*, *Cryptantha*, and *Amaranthus* could not be identified to species with any confidence and were treated at the generic level. Both *Schismus arabicus* and *S. barbatus* were present in the seed bank, sometimes in the same sample, but were treated as *Schismus* sp. because seeds that had fallen from the florets could not be reliably identified to species. Identified seeds were

tested for viability by cutting them in half with a razor blade, then inspecting them. Firm, plump, undamaged seeds with moist, fleshy, or oily embryos were considered viable; seeds with discolored, crumbly, chalky, shrunken, or missing embryos were deemed nonviable and discarded. For each sample, a running tally was kept of viable seeds by species.

Sawma and Mohler (2002) provide a brief discussion of the advantages and disadvantages of various tests for seed viability. One method involves germinating seeds in flats, then counting and identifying seedlings as soon as possible after emergence. Another is to cut seeds in half, moisten the cut surface with a tetrazolium solution, then place seeds at a suitable temperature and watch for the characteristic red staining of respiring tissues. Tetrazolium testing requires a temperature-controlled environment and is laborious when small seeds or many seeds must be tested. Germination testing requires a greenhouse or shadehouse with automatic irrigation equipment and, because some dormant seeds might never emerge as seedlings, tends to underestimate the viable seed bank. Visual inspection of cut seeds is less labor-intensive than the tetrazolium method and less apt to overlook dormant seeds than the germination method. Although visual inspection might pass some nonviable seeds as viable, it is a useful and efficient technique for seed-bank surveys (Sawma and Mohler 2002).

Wilcoxon Rank Sums tests were used to compare samples from 2006 and samples from untreated areas in 2007 (burned and mowed samples were excluded to remove bias) in terms of number of seeds dm^{-2} , number of species dm^{-2} , number of native seeds dm^{-2} , number of exotic seeds dm^{-2} , number of native species dm^{-2} , and number of exotic species dm^{-2} . Similarly, Wilcoxon Rank Sums tests were used to compare treatments in the 2007 samples. 40 samples were collected in June 2006 and 45 samples (15 samples each for burned, mowed, and untreated areas) were collected in July 2007. All means are reported as ± 1 SD.

Results

A total of 36 species were found in this study, with all but 4 species (*P. ciliare*, *E. lehmanniana*, *Sporobolus airoides*, and *Porophyllum gracile*- 3 grasses [2 invasive exotics] and 1 subshrub) being annuals. A total of 22 species were found in 2006 and 24 species were found in 2007.

From June 2006 to July 2007 (Table 2), the density of viable seeds dm^{-2} declined from 62.2 to 18.8, with native species declining from 25.9 to 16.1 viable seeds dm^{-2} and exotic species declining from 36.2 to 2.7 viable seeds dm^{-2} . Species richness also declined from June 2006 to July 2007, with the number of species dm^{-2} falling from 5.5 to 2.8. Native species richness declined from 3.5 to 1.9 species dm^{-2} and exotic species richness declined from 2.0 to 0.9 species dm^{-2} . *P. ciliare*, *Schismus* sp., *Amsinkia menziesii*, *Descurainia pinnata*, *Euphorbia* sp., *Plantago* sp., and *Salsola kali* all declined from 2006 to 2007, with *P. ciliare* and *Schismus* showing the most dramatic declines from 10.6 to 1.8 seeds dm^{-2} and from 24.4 to 0.9 seeds dm^{-2} , respectively. Conversely, the number of seeds of *Boerhavia* sp., *Bouteloua aristidoides*, *Pectocarya recurvata*, and an unidentified species (Unknown 3) all increased from 2006 to 2007, with the most dramatic increase occurring for *B. aristidoides* which increased from 0.1 to 5.1 seeds dm^{-2} .

Among the different treatments applied in 2007 (Table 3), no significant differences were found in the density of viable seeds dm^{-2} overall, or in the density of either viable native or exotic seeds dm^{-2} . Similarly, no significant differences were found in the species richness dm^{-2} overall, or in the species richness of native species dm^{-2} . However, the number of exotic species dm^{-2} was slightly higher in the burned (1.6 species dm^{-2}) than in the untreated (0.9 species dm^{-2})

samples. *Schismus* sp. seed densities in the burned (4.7 seeds dm⁻²) samples were higher than the untreated (0.9 seeds dm⁻²) samples. *A. menziesii*, *Erodium cicutarium*, and *Euphorbia* sp. seed densities were higher in the mowed samples than in the burned or untreated samples. *P. recurvata* seed densities were higher in the untreated samples than in the burned or mowed samples and Unknown 4 seed densities were higher in the burned samples than in the mowed or untreated samples.

Discussion

Table 2 shows the differences between seed densities and species richness from June 2006 to July 2007. A dramatic decline is evident in all major species, especially the two most abundant species, *Schismus* and *P. ciliare*, which accounted for over 56% of the seedbank in 2006. Several potential explanations for the decline exist. First, a large portion of the 2006 seedbank may have germinated after summer 2006 or spring 2007 rains but failed to persist, thus depleting the seedbank. Another possibility is that the soil surface of the former agricultural fields hold relatively few safe sites for seeds (as opposed to a natural surface that would have many nooks and crevices among the rocks and minor soil topography) and most seeds were either blown away by strong wind or washed away by sheet flow. Granivory by rodents or insects could be another explanation (Price and Joyner 1997).

Further complicating this dramatic decline in seedbank densities is the fact that 2006 was officially an El Niño year (<http://www.wrcc.dri.edu/enso/ensodef.html>), which should have resulted in high seed production for any warm season flowering species (most notably *P. ciliare*). The seed resulting from this high production event might have met a fate similar to that of the seed already in the soil at the time of the 2006 sampling. Contrary to the assumption stated in

the introduction (question 2), if seed production was increased due to the El Niño event, this did not translate to seed presence in the seedbank, which has strong implications for natural and assisted recovery, i.e. conditions producing large amounts of seed (either direct seeding or climatic conditions that lead to high seed production in existing plants) will not necessarily produce increases in the seedbank or in plant establishment. Further research would help elaborate on the significance of this finding to restoration activities (will the fate of seeds broadcast or drilled be the same as seeds produced naturally- both disappearing from the site without resulting in seedling emergence and subsequent establishment?).

Table 3 indicates few apparent differences in the seedbank under different buffelgrass treatments, and where differences did occur, they were small in magnitude, especially compared to the differences observed between years sampled. A couple of cases are worth noting, however. *Schismus* seed densities were over 5x higher in burned samples than in untreated samples. This points to the adaptation of *Schismus* to fire and suggests that fire would be an inappropriate control technique for this short-lived annual. The small increase in density of exotic species in the seedbank in burned versus mowed or untreated plots can likely be attributed to the increase in *Schismus* in these plots. Also interesting is that *P. ciliare*, also a fire-adapted invasive species, showed no difference in seed densities between burned and mowed or untreated areas. This may be due to the fact that *P. ciliare* is a perennial and therefore relies not on the seedbank for persistence through fire but on resprouting from mature plants.

Conclusions

Though a brief snapshot in time, this study already demonstrates the enormous temporal and spatial variability typical of desert seed banks (Reichman 1984, Kemp 1989, Guo et al.

1999). We were unable to answer the questions posed in the introduction because no treatments were performed during the 2 years we sampled. Continuing the study into the future will greatly increase our ability to answer the basic questions posed about 1) the length of time buffelgrass will persist in the seedbank given that control treatments continue and 2) the changes in the native seedbank as control efforts continue. At present, we are limited in making inferences about the effects of various treatments on the seedbank. We plan to sample again prior to the 2008 monsoon and would like to continue sampling in future years, at least till 2010 to ensure 3 years post treatment data, as suggested in the original proposal (Appendix A). If COT agrees to continue the project, we recommend that the schedule be amended to reflect that the first year of post herbicide treatment collections be made in 2008.

Although the original questions could not be addressed due to circumstances outside our control, this brief study nonetheless yielded an interesting result- a drop, not an increase, in seedbank densities following an El Niño event. We suggest further investigations into this phenomenon using a combination of seed traps and collecting seedheads on plants to estimate seed production and to measure how much seed is actually reaching the soil on site before being predated or dispersed off site. If most seed produced is reaching the soil but is not showing up in the seedbank or as established plants, it is either germinating and dying, being killed by extreme climate conditions (heat/sunlight), or being predated by granivores. Any of these scenarios would present a significant obstacle to restoration using seed. Another interesting study would involve initiating a seed rain/seedbank study on lands being actively seeded by COT as a restoration activity to determine the fate of applied seeds. We would very much like to explore this idea further and would be happy to present a research proposal to COT if desired.

Acknowledgements

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Figure 1: February 2007 map of buffelgrass infestation at COT property near Manville and Reservation Roads in Avra Valley, Arizona, showing areas where seedbank samples were collected. Red line indicates the boundary of the mapped area, buffelgrass stands, yellow indicates the infested areas, and the cross-hatching indicates areas that were mowed (preventing accurate mapping).

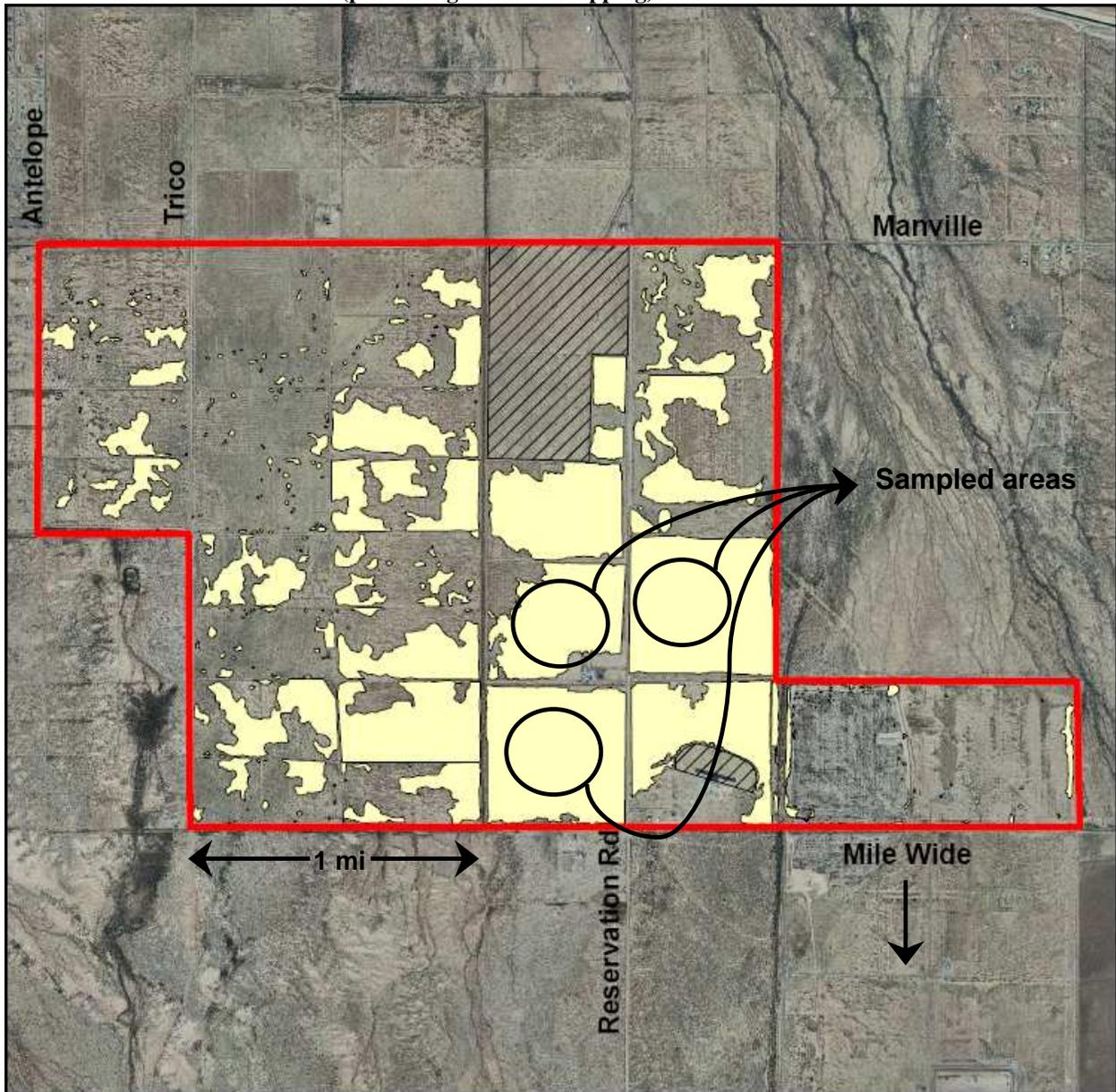


Table 1: Timeline of buffelgrass treatment activities at COT property in Avra Valley, Arizona.

Date	Treatment	Acres
February 2007	Mowed/Burned	400/400
May 2007	Sprayed with herbicide	80
July-September 2007	Sprayed with herbicide	1200
April-May 2008	Mowed	900
May 2008	Burned	160

Table 2: Mean seed densities (seeds dm⁻²) and species richness (# species) of samples taken in 2006 and 2007 on the COT property in Avra Valley, Arizona. Burned and mowed treatments were excluded from 2007 data.

Species	2006 (n=40)		2007 (n=15)	
	Mean	SD	Mean	SD
<i>Pennisetum ciliare</i> *	10.6 ^a	12.8	1.8 ^b	3.6
<i>Schismus arabicus</i> , <i>S. barbatus</i> *	24.4 ^a	19.4	0.9 ^b	1.3
<i>Amaranthus</i> sp.	0.1	0.3	0	0
<i>Amsinckia menziesii</i>	3.2 ^a	6.6	0 ^b	0
<i>Amsinckia tessellata</i>	0.1	0.2	0	0
<i>Aristida adscensionis</i>	0	0	0.2	0.8
<i>Boerhavia</i> sp.	0 ^b	0	0.5 ^a	1.4
<i>Bouteloua aristidoides</i>	0.1 ^b	0.5	5.1 ^a	13.4
<i>Bromus rubens</i> *	0.0	0.2	0	0
<i>Cryptantha angustifolia</i>	0.4	1.3	0	0
<i>Cryptantha barbigera</i>	0.0	0.2	0	0
<i>Cryptantha pterocarya</i>	0.0	0.2	0	0
<i>Cryptantha</i> sp.	0.0	0.2	0	0
<i>Daucus pusillus</i>	0.0	0.2	0	0
<i>Descurainia pinnata</i>	1.8 ^a	6.0	0 ^b	0
<i>Eriophyllum lanosum</i>	0.1	0.6	0	0
<i>Euphorbia</i> sp.	2.3 ^a	3.5	0.1 ^b	0.4
<i>Lappula occidentalis</i>	0.2	0.7	0	0
<i>Lepidium</i> sp.	0.2	0.8	0.1	0.4
<i>Mentzelia</i> sp.	0.4	1.2	0	0
<i>Pectocarya heterocarpa</i>	3.1	10.0	2.4	9.0
<i>Pectocarya platycarpa</i>	1.5	7.2	3.3	12.9
<i>Pectocarya recurvata</i>	1.4 ^b	7.9	2.1 ^a	5.1
<i>Plantago</i> sp.	7.2 ^a	12.5	1.7 ^b	4.9
<i>Porophyllum gracile</i>	0.1	0.3	0	0
<i>Salsola kali</i> *	1.2 ^a	2.6	0 ^b	0
<i>Stylocline micropoides</i>	3.8	12.3	0	0
Unknown 1	0	0	0.1	0.3
Unknown 2	0	0	0.1	0.3
Unknown 3	0 ^b	0	0.2 ^a	0.6
Unknown 4	0	0	0.1	0.3
Unknown 5	0.1	0.4	0	0
Unknown 6	0.1	0.3	0	0
All seeds	62.2 ^a	48.2	18.8 ^b	37.8
All species	5.5 ^a	2.5	2.8 ^b	2.5
Native seeds	25.9 ^a	38.1	16.1 ^b	34.6
Native species	3.5 ^a	2.0	1.9 ^b	2.1
Exotic seeds	36.2 ^a	24.3	2.7 ^b	3.8
Exotic species	2.0 ^a	0.8	0.9 ^b	0.7

Means followed by different letters within a row were determined to be significantly different at $p < 0.05$ using Wilcoxon Rank Sums tests. An asterisk indicates a species not native to North America.

Table 3: Mean seed densities (seeds dm⁻²) and species richness (# species) of burned, mowed, and untreated samples taken 2007 on the COT property in Avra Valley, Arizona.

Species	Burned (n=15)		Mowed (n=15)		Untreated (n=15)	
	Mean	SD	Mean	SD	Mean	SD
<i>Pennisetum ciliare</i> *	0.9	1.9	1.1	2.0	1.8	3.6
<i>Schismus arabicus</i> , <i>S. barbatus</i> *	4.7 ^a	4.5	2.7 ^{ab}	5.7	0.9 ^b	1.3
<i>Amaranthus</i> sp.	0	0	1.1	4.4	0	0
<i>Amsinckia menziesii</i>	0 ^b	0	0.3 ^a	0.6	0 ^b	0
<i>Amsinckia tessellata</i>	0.1	0.3	0	0	0	0
<i>Aristida adscensionis</i>	0	0	1.9	5.2	0.2	0.8
<i>Boerhavia</i> sp.	0	0	0	0	0.5	1.4
<i>Bouteloua aristidoides</i>	16.5	19.1	4.6	8.1	5.1	13.4
<i>Descurainia pinnata</i>	0	0	0.1	0.5	0	0
<i>Eragrostis lehmanniana</i> *	0.2	0.6	0	0	0	0
<i>Erodium cicutarium</i> *	0 ^b	0	1.5 ^a	4.9	0 ^b	0
<i>Euphorbia</i> sp.	0.7 ^b	1.5	2.8 ^a	2.4	0.1 ^b	0.4
<i>Lepidium</i> sp.	0	0	0	0	0.1	0.4
<i>Mentzelia</i> sp.	0	0	0.1	0.3	0	0
<i>Pectocarya heterocarpa</i>	0.5	1.8	0	0	2.4	9.0
<i>Pectocarya platycarpa</i>	0	0	0	0	3.3	12.9
<i>Pectocarya recurvata</i>	0 ^b	0	0 ^b	0	2.1 ^a	5.1
<i>Plantago</i> sp.	0	0	0.3	1.0	1.7	4.9
<i>Salsola kali</i> *	0.1	0.4	0	0	0	0
<i>Sporobolus airoides</i>	0.1	0.5	0	0	0	0
Unknown 1	0.1	0.1	0.1	0.1	0.1	0.1
Unknown 2	0	0	0	0	0.1	0.3
Unknown 3	0.1	0.4	0.1	0.3	0.2	0.6
Unknown 4	0.9 ^a	1.4	0 ^b	0	0.1 ^b	0.2
All seeds	24.9	22.6	16.7	17.8	18.8	37.8
All species	3.6	1.6	3.2	1.9	2.8	2.5
Native seeds	19.0	19.6	11.5	13.3	16.1	34.6
Native species	2.0	01.1	2.3	1.5	1.9	2.1
Exotic seeds	5.9	5.7	5.2	8.4	2.7	3.8
Exotic species	1.6 ^a	0.7	0.9 ^{ab}	0.9	0.9 ^b	0.7

Means followed by different letters within a row are significantly different at $p < 0.05$ by the Wilcoxon Rank Sums tests. An asterisk indicates a species not native to North America.

Appendix A: Original proposal to COT for seedbank work

Native and buffelgrass seedbank characteristics following chemical control of buffelgrass on City of Tucson property in Avra Valley

Submitted to COT HCP TAC by Travis M. Bean on 24 July 2006

Buffelgrass is the dominant vegetation on some City of Tucson (COT)-owned properties near Reservation Rd and Mile-Wide Rd in Avra Valley. Buffelgrass is widely recognized as a serious threat to life, property, biodiversity, and local economies in the Sonoran Desert. COT will soon begin to use herbicides to control buffelgrass on these properties to 1. lessen the risk of catastrophic fire that could spread to nearby private inholdings, 2. reduce the seed source of buffelgrass in the area to prevent its continued spread into nearby Saguaro National Park and Tucson Mountain Park, and 3. facilitate the recovery of native vegetation on this property. This study is relevant to items 2 and 3.

By reducing the density of live buffelgrass plants (and therefore buffelgrass seed production) on COT Avra Valley properties, we hope that the seedbank density of buffelgrass will also become less dense and will decrease in viability over time. Similarly, because we assume that buffelgrass densities are sufficient to limit establishment and/or persistence of some native species, and that these species may begin to recolonize following reduction in density of live buffelgrass plants, we hope that native seedbank density and viability will increase over time.

The two basic questions we hope to answer are:

1. how long does buffelgrass seed remain viable in the soil?
2. will buffelgrass seed in the soil be replaced by native seed as buffelgrass seed inputs are (presumably) reduced?

Both questions can be addressed by examining the soil seedbank on the property. In June 2006, prior to monsoonal rains that would trigger buffelgrass green up (and chemical control) and seed production, we took 40 soil samples in the buffelgrass infested area near Mile-Wide and Reservation Roads. Four sites were randomly located in the area and 10 samples were collected from random locations at each site. This degree of replication should be sufficient given the homogeneity of the plant community in the immediate area of the buffelgrass infestation. Samples will be taken again after seed has shattered (dropped from parent plants) in Sept-Oct in this year and each of the next three years (2006-2009).

Samples will be analyzed for density of viable buffelgrass and native seed through time. We hope this information will allow us to estimate the longevity of buffelgrass seed in the soil at this site and to document any changes in native seed density following chemical control of buffelgrass. A suggested schedule and budget are on page 2.

Please direct any questions, comments or concerns to Travis Bean (bean@email.arizona.edu, 629.9455x104).

Date	Activity
June 2006	collect baseline soil samples
September-October 2006	collect first year post-spraying samples
October 2006-August 2007	analyze samples
August 2007	annual report to TAC
September-October 2007	collect second year post-spraying samples
October 2007-August 2008	analyze samples
August 2008	annual report to TAC
September-October 2008	collect third year post-spraying samples
October 2008-August 2009	analyze samples
August 2009	final report to TAC

Budget

		FY 07	FY 08	FY 09	FY 10	TOTAL
student employee (2 people, \$10/hr, 230 hrs*)	collection, flotation, sorting of soil seedbank samples	\$4,600	\$2,300	\$2,300	\$2,300	\$11,500
ERE student workers (3.6%)		\$166	\$76	\$76	\$76	\$394
Total salaries, wages, and benefits		\$4,766	\$2,376	\$2,376	\$2,376	\$11,894
Travel	(40 mi roundtrip, 2 trips 1st year, 1 trip/yr thereafter, \$0.405/mi)	\$30	\$15	\$15	\$15	\$75
Total direct costs		\$4,796	\$2,391	\$2,391	\$2,391	\$11,969
Indirect costs (K * 0.26)		\$1,247	\$622	\$622	\$622	\$3,112
Total direct and indirect costs		\$6,043	\$3,013	\$3,013	\$3,013	\$15,081

*20 person-hrs to collect samples, 50 hrs to float samples, 160 hrs to sort samples = 230 person-hrs (x2 in yr 1)