

2020

Supplemental seeding and seed predation in a newly planted tallgrass prairie

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Recommended Citation

Betzer, Isabella, "Supplemental seeding and seed predation in a newly planted tallgrass prairie" (2020).
Honors Program Theses. 447.
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SUPPLEMENTAL SEEDING AND SEED
PREDATION IN A NEWLY PLANTED TALLGRASS PRAIRIE

A Thesis Submitted
in Partial Fulfillment
of the Requirements for the Designation
University Honors

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November 2020

This Study by: Isabella Betzer

Entitled: Supplemental Seeding and Seed Predation in a Newly Planted Tallgrass Prairie

has been approved as meeting the thesis or project requirement for the Designation

University Honors

Date

Dr. Laura Jackson, Honors Thesis Advisor

Date

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Abstract

The Iowa around today looks vastly different from the Iowa that was originally around. What once used to house vast tallgrass prairies is now home to about 0.1% of what was originally here (Smith 1998). However, conservationists have been working hard to restore even a fraction of what originally here despite high costs and low yields in restoration prairies. Recently experiments have been done on the use of sacrificial seed to potentially increase plant yield in restoration with successful results. This study was done to further research the effects of supplemental seeding on seed predation and if seedling emergence would increase with supplemental seeding. Research on predation was done using seed cards and seedling emergence was counted after three months of growth. Results found that there was no change in seed predation or seedling emergence between control plots and plots containing supplemental seed. Further research in the area of supplemental seeding is necessary to determine the full capabilities of this method.

Acknowledgments

Thank you to Dr. Laura Jackson my thesis advisor who provided her guidance and expertise throughout the process of researching and writing. Thank you to Jacey Meier, a fellow biology student at the University of Northern Iowa, for her assistance in the field and during data analysis. Thank you to the Tallgrass Prairie Center at the University of Northern Iowa and the University of Northern Iowa Honors Program for their support.

The Iowa that is here today is not the Iowa that was originally discovered. Looking into pre-settlement Iowa, it is estimated about 28.6 million acres of tallgrass prairie originally covered the state, which translates to 79.45% of the state (Smith 1998). These prairies, mixed with the other ecosystems of Iowa, produced a colorful landscape of complex ecological interactions (Smith 1998). In fact, prairies have been a part of Iowa's natural landscape for an exceptionally long time. Pollen records from the past 30,000 years show that Iowa's landscape has shifted between coniferous forests, deciduous forests, and prairies (Smith 1981). These numbers obviously do not represent the lands around today.

Ever so slowly, native prairie lands have been dwindling down in numbers. Outside of Iowa, it is estimated that of the original 240 million acres of tallgrass prairie only about 3% remains (Smith 1992). This loss is even greater when looking at Iowa, by 1920 Iowa retained 1.7% of its natural prairie and today it is estimated that Iowa retains less than 0.1% of its natural prairie (Smith 1998). These low numbers go even lower when talking about remaining prairies containing similar species composition to pre-settlement prairies, going from less than 0.1% to less than 0.05% (Smith 1998).

Prairies are a unique and complex ecosystem that provide a number of different benefits including: native habitats for wildlife, erosion control, increased water absorption in soil, invasive weed control, carbon sequestration, and nutrient reduction (Tallgrass Prairie Center 2020-a). Some of these benefits are greatly important in today's climate, including erosion control and carbon sequestration. Today's Iowa is dominated by row crops which provides inadequate protection for topsoil, but perennial prairie roots provide the protection and support that topsoil needs to prevent runoff (Tallgrass Prairie Center 2020-a). Mature undisturbed prairies can also

provide a deep well of storage for carbon in their roots below ground, more than forests can store above ground (Tallgrass Prairie Center 2020-a).

It is no question that Iowa has lost a vast majority of its native landscape, but that does not mean that efforts are not being made today to help restore even just a little of what has been lost. Conservation efforts have been around since the late 1800's and have continued to today (Smith 1998). Even here at the University of Northern Iowa (UNI) there are conservation efforts being made. The Tallgrass Prairie Center (TPC) at UNI puts a lot of focus into the research and restoration of tallgrass prairies in Iowa (Tallgrass Prairie Center 2020-b). The TPC strives to, “improve our understanding of prairie reconstruction and ecology while at the same time put more prairie back on the landscape to benefit people, the land, and other living things” (Tallgrass Prairie Center 2020). However, there are many roadblocks that stand in the way of prairie restoration.

One thing standing in the way of prairie restoration are the high costs. Native seed banks are largely depleted, making commercial seed the main source of seeds for prairie restoration (Smith 1998). This is a very costly path, one example being the Glacial Ridge prairie restoration project. During the Glacial Ridge prairie restoration project about \$8.2 million was spent on direct restoration, and of this more than half the money went towards the purchasing and harvesting of seeds required for a high-diversity planting (Gerla et. al. 2012). Commercial seed can cost anywhere from \$963/acre to \$2,819/acre depending on the seed mix, with lower diversity mixes usually costing less than high diversity mixtures (Prairie Moon Nursery 2020). These high prices are further inflated by the number of seeds needed for desired plant density. To achieve a density of about 30 adult plants/m² a planting must have between 400 to 950 pure live

seeds/m² (an establishment rate of 3.1-7.5%) (Smith et. al. 2010). These low rates of seedling establishment mean high costs for little yield. So what factors cause this?

One factor of low establishment can be attributed to seed predators, otherwise called granivores. Granivores are species that feed on seeds and grains and are abundant among Iowa's, and the world's, ecosystems. It is well archived that granivores, such as birds and rodents, not only prey on prairie restorations but can also shape the overall composition of plant communities (Howe & Brown 1999, 2000; Pellish et. al. 2017; Westerman et. al 2003). Although predation is not the only factor in low seedling establishment rates, small vertebrate granivores have been shown to reduce seedling emergence rates by 5% and can reduce the total number of emerging seedlings by about 30% (Pellish et. al. 2017). Given this impact, there is importance in the research of methods that can help reduce the impact of granivores on prairie restoration.

Fighting these low establishment numbers might be as simple as looking towards nature and the phenomenon known as mast seeding (often referred to as supplemental seeding in prairie restoration). Mast seeding is the synchronous production of large seed crops in plants (Kelly 1994 & 2002). Masting is an evolutionary tactic that is still being studied today, but one theory for why some plants do this is the predator satiation theory. In order to ensure the continuation of a species, seeds must survive a number of factors to germinate, one of these being their predators. The predator satiation theory suggests that large intermittent seed crops reduce seed losses to their predators (Janzen 1971; Kelly 2000 & 2002).

This theory is rather widely accepted, but its effectiveness is dependent on predator interactions (Kelly 2002). All of this has to do with plants already established in their own ecosystem, but the transferring of the idea of mast seeding to prairie restoration has shown potential in increasing the seedling establishment. In roadside prairie restoration, while not

decreasing the rates of seed consumption, the inclusion of supplemental seed has been shown to increase seedling establishment by approximately 37% (Riebkes et. al. 2018). In this study further exploration of the use of supplemental seeding is used to answer the following questions: (1) Does supplemental seeding effect seed predation by granivores in a newly planted prairie? and (2) Will supplemental seeding increase seedling establishment in prairie restoration? Based on previous research it was predicted that the predation would be difficult to track between supplemental seeding, and that seedling establishment would increase for plots with supplemental seed.

Methods

The research for my thesis was conducted in the spring and summer of 2020 at the Irvine prairie. The Irvine prairie is a 77-acre area located in Benton County, Iowa that was donated by Cathy Irvine in 2018 to be used as a site for prairie restoration. The area started as row crop farmland and is in the ongoing process of being restored. The 18.5-acre 2020 section was used as the basis for this research. The 2020 plots were seeded on March 31 and April 2 using a seed mix similar to that of the 2019 Irvine prairie planting (Meissen 2020). The prairie seed was planted at 400 seeds per square meter following similar methods as the 2019 Irvine prairie planting (Meissen 2020). On April 2 six adjacent 40m² plots were designated using stakes and the supplemental bird seed was distributed on April 9 across three of the six plots. Supplemental bird seed was planted at 10x the weight of the prairie seed planted. The plots without supplemental seeding, plots 2, 3 and 6, acted as control plots for the experiment. On April 23, these plots were then further divided into 10m² plots at the center and these 10m² plots were marked with flags and the basis of the seed card research. Figure 1 shows a recreation of the plots. The prairie was mowed four times during the summer, however, the 10m² plots were mowed around.

Seed Predation

The model for our seed cards were reproduced from the Westerman et. al. (2003) paper. Seed cards were made using 21.6 cm x 27.9 cm sheets of sandpaper (3M Paper Sheet 346U, 36 Grit). The sheets were torn into roughly equal fourths and then gridded into fourths using black markers to help in the counting process. For the seeds, we decided to use *Heliopsis helianthoides*. These small black seeds were already a part of the seed mix and their size and shape made for easy distinction on the sandpaper cards. Seeds were counted by hand and placed into vials for easy application onto the seed cards. For application aerosol glue (3M Super Multipurpose Adhesive Aerosol) was sprayed onto the card, seeds were applied, and then another layer of glue was applied. Glue does not prevent predation on seed cards and has not been shown to have any ill effects on the granivores (Westerman et. al. 2003). Cards were made at least 24 hours prior to field application to allow for drying. At the beginning of the field application and data collection there was a distinct lack of seeds, viable or not, left on the cards upon their collection. Trial 3 was counted twice to assess if this was due to gluing application or time. It was determined from trial 3 that the method of glue application was not at the necessary level to prevent seed loss from weather related loss (wind, rain, etc.). To counteract this, the method of gluing was changed, starting during trial 4, to apply a thicker coating, which still allowed for seed loss by predators but helped in the prevention of weather-related losses. In addition to heavier glue application, the timing in between counting was shortened to record shorter predation times.

Seed cards were first distributed in the plots on April 23, 2020. The 10m² plots were gridded and then four random points along the one, three, five, seven, and nine-meter mark were selected using a random number generator. These points were used over the course of the testing time for

ease of application. The cards were secured to the ground using two 5.1 cm, flat-top roofing nails at opposite corners. The nails were pushed through the card and then pushed into the ground so that the card laid as flat as possible. Seed cards were counted, recorded, and replaced with new cards roughly every two weeks for the first two trials, starting with the third trial cards were counted a week after instillation and replaced every two weeks. Due to weather conditions cards could not always be counted at exactly on schedule. Any seeds on the seed card that were deemed unviable (broken or partially eaten) were not included in the seed count. Table 1 shows the dates of instillation and counting for all six trials conducted.

Trial Number	Date Installed	Date Counted
1	4-23-2020	5-14-2020
2	5-14-2020	5-29-2020
3	5-29-2020	6-4-2020 & 6-12-2020
4	6-12-2020	6-20-2020
5	6-25-2020	7-2-2020
6	7-9-2020	7-16-2020

Table 1 – Installation and Counting Dates

Control Cages

The control cages were used to compare the seed loss by non-predators (ex: weather) versus predators. However, the process of developing a control cage was an endeavor that changed as the experiment went on. The first round of cages was created using a plastic base that raised 20cm off the ground. The base was 63cm by 34cm with holes along the bottom. A dome of gridded wire was attached using zip ties. The wire used for the first batch of cages had 7mm

squares. The ends and bottom of the base were then also covered with 3mm² gridded wire cut to fit the opening, these were also attached using zip ties, allowing for the opening of the cages when changing control cards. Eight control cards were then stuck to the base using duct tape on the bottom of the cards to allow for easy removal by hand, but also to prevent the cards from moving around in the cages. For field application, the cages were placed at a random point on the edges of the 10m² area used for experimentation. To keep the cages in place, two long pole stakes were affixed through the cages. These cages were applied May 29.:

It was found that these cages were ineffective in their design to exclude all predators. Small invertebrates were seen inside the cages, meaning instead of excluding all seed predators they were only effective in excluding mammalian predators and underground predators. The second round of cages were created using the original cages with a modified design. The dome around the cage was covered by an additional layer of gridded wire consisting of 3mm squares. To further help in the prevention of nonvertebrate predators, two sticky traps were affixed inside the cage using duct tape on the bottom of the traps at the time that the control cards were placed inside, and tanglefoot was applied on site to the legs of the cages prior to their placement on the field. Also, upon application to the field any plants that was hanging around the cages was trimmed to further prevent any entry points for nonvertebrate granivores. These upgraded cages were installed July 9 and showed a significant difference in the number of seeds counted on the control cards.

Seedling Establishment

In order to test if the supplemental seeding had an effect on seedling emergence, two seedling counts were conducted on July 14 and 17. Two random directions (North, South, East, West, Northwest, Northeast, Southwest, and Southeast) were selected for each plot using a

random number generator. From these directions a 10m x 10m plot of land was sectioned off, as can be seen in fig 1. Ten random coordinates were generated for each plot. A 0.25 m² metal frame was used at each coordinate to count seedlings into two categories: native forbs and native grasses. Prior to counting, those of us involved were given a lesson in identifying native grasses and forbs. Books on native plant identification were also used to help in the identification of the seedlings (Williams 2010). Due to the nature of young native grasses being hard to identify, grass identification was harder to quantify versus the native forbs.

Data

Seed Predation

A total of six trials were performed over the course of April 2020 to July 2020. For each trial, the percentage of seeds removed from seed cards was calculated. Due to ineffective gluing methods, the first three trials have been excluded from data analysis. Trials 4 and 5 were calculated as uncontrolled due to the control cages ineffectiveness against invertebrate predators, and trial 6, using the new cages, was calculated controlled. To correct for controls, the amount of seeds left on the seed cards was subtracted from the average of the exposed seeds left on the control cards. The total was then divided by the average of the seeds left on the control cards. Lastly, the mean percentage of seeds removed from seed cards was calculated for each plot for trials 4, 5, and 6 along with the standard error (SE).

Seedling Establishment

To assess seedling establishment, we started by adding up the total number of native forbs and native grasses counted for each plot for both days of counting. These numbers were then averaged to find the average number of native forbs and native grasses per sq meter for each plot.

Both sets of data, for the seed card predation and seedling establishment, were plotted using bar graphs in sigma plot version 14.

Predator Exclusion

Although not a part of the original experiment, the mean percentage and standard error of seeds removed from exposed seed cards, control seed cards from trial 4 and 5, and control seed cards from trial 6 were calculated to find a rough trend in the removal of seeds at different levels of exclusion (no exclusion, partial, and complete). Data was plotted as a bar graph in sigma plot version 14.

Results

Seed Predation

For the seed predation we were unable to track any significant difference between treated plots and control plots (Figure 2). This followed for all the trials used (trials 4, 5, and 6), and suggests that any difference in predation was unable to be tracked by the methods used. The mean percentage of seeds removed for the control plots found were: 93.66% (± 3.61 SE), 92.78% (± 4.04), and 93.46% (± 3.77). For the treated plots, the mean percentages found were: 91.33% (± 2.43), 89.44% (± 3.01), and 90.88% (± 2.61).

Seedling Establishment

Seedling emergence between treated and control plots were similar and no statistically significant difference was found between the two (Figure 3). This suggest that the supplemental seeding application had no visible impact on the seedling emergence. On average, for the control plots, 14.7 native forbs/m² and 5.67 native grasses/m² were established at the time of the

experiments end. For the treated plots, an average of 15.3 native forbs/m² and 7.33 native grasses/m² were established at the time of the experiments end.

Predator Exclusion

As mentioned previously, the mean percentage of seed removal was calculated on three different levels of exclusion: no exclusion, partial exclusion, and complete exclusion. Figure 4 shows these levels, no exclusion had 91.93% (± 0.43) removal, partial exclusion had 57.19% (± 3.01) removal, and complete exclusion had 3.68% (± 0.85) removal. Notice the large differences in n due to the data analyzed not being a part of the original experiment.

Discussion

We wanted to examine the impact of supplemental seeding on seed predation and seedling establishment in a newly planted prairie restoration. This was done by examining seed predation from seed cards, and mid-summer seedling emergence in treated and control plots. Another factor of predator exclusion was examined using the data from the seed predation experiment. Seed card data showed that seed predators were able to remove large numbers of seeds from an area, but there was no evidence found from the seedling data that the application of supplemental seed reduced the consumption of prairie seeds by seed predators. These results suggest that supplemental seeding in April did not significantly impact rates of seed predation or seedling emergence. Based on previous research (Riebkes et. al. 2018) we predicted that any seed predation difference would be hard to track.

Although our results on seedling emergence do not show the predicted results, seed predators still retain an impact on prairie restoration, costing an estimated \$180 to \$250 per hectare (Pellish et. al. 2017). There is still important research to be done on the potential impact

of supplemental seeding that was not able to be done in our experiment. Prairie restoration is a lengthy process, and it can take years for a new prairie to reach full growth and diversity. What was counted on 6/25/2020 and 7/9/2020 was incomplete, leaving out important grasses we were unable to identify, and later plant establishment in July and August. So, due to the short timeline of the experiment and inexperience in grass identification, the true impact that supplemental seeding might have not been measured. Further counts and experimentation might show differing results for seedling emergence.

Furthermore, in this experiment supplemental seed was applied only once at the beginning of the experiment, several days after the prairie was seeded. This time allowed for a period of predation without supplemental seed. As soon as the seed was out starlings could be seen throughout the field eating what was available, and this is just visible predation. Predation happens in several ways including those that are visible, such as birds and small mammals, and those that are invisible, underground predation. Earthworm middens were very abundant throughout the prairie, including the study area, and the effects of earthworms on seedlings is well documented (Milcu et. al. 2006). Although any impact this gap of time might have had is unknown, future research could allow for the investigation into any impact it might have had. It would also be interesting to investigate how the application of supplemental seed over time may impact seedling emergence, spreading the supply of supplemental seed over a period to allow for establishment of native seeds.

Further research could also allow for more experimentation into the best method to track seed predation in supplementally seeded prairie restorations. In both our research and the Riebkes et. al. (2018) paper it was found that tracking any seed predation differences between control and treated plots was difficult. With the understanding of how masting works and the

predator satiation theory it would make sense that predation between the control plots and treated plots would change as predators are able to eat their fill from the treated plots. We may have been too late in tracking seed predation to have found any difference between treated and control plot. Our first seed card trial that we were confident in happened in June while the supplemental seed was added in April. This missed time may have been important in tracking the full picture of seed predation in between our plots.

Prairie restoration is an important cause, and there are many benefits that can be gained through it such as native habitats for wildlife, erosion control, increased water absorption in soil, invasive weed control, carbon sequestration, and nutrient reduction (Tallgrass Prairie Center 2020-a). It is important that research continues to find ways to help cut the costs that are associated with prairie restoration, and supplemental seeding is one potential avenue for this. Although we were unable to recreate previous findings that does not mean that the full impact of supplemental seeding is understood. Further research could help broaden the understanding of how supplemental seeding works and its impact on predation in prairie restoration.

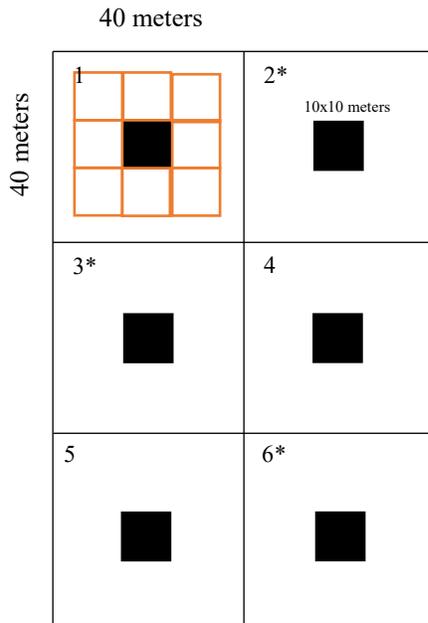


Figure 1 - Plots with * denote supplemental seeding. Black squares denote 10x10 sections containing seed cards. Orange squares denote 10x10 sections for seedling counts.

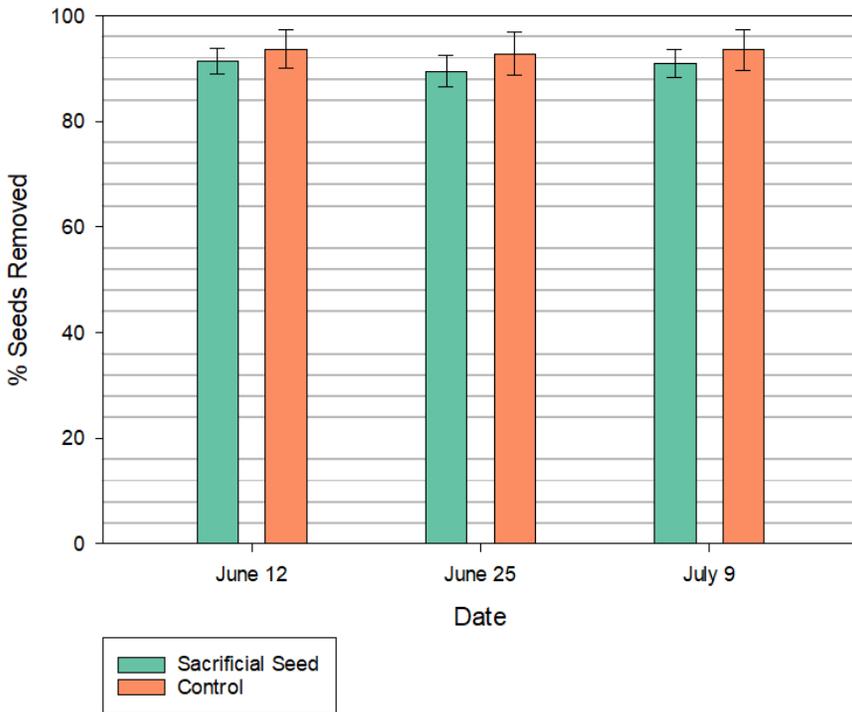


Figure 2 – Percentage of seed removal from seed cards by predators in a planted prairie. One hundred and twenty seed cards made using *Heliopsis helianthoides* were placed in six plots and counted by hand after a week of exposure. Control cages containing eight seed cards were placed at each plot June 12 and 25. Controls were protected from mammals and birds and the July 9 controls were protected from all predators. Results show no significant difference between treated plots and control plots but show definitive predation of seeds in the prairie. Data shown as mean percentage of seeds removed from seed cards by predators adjusted for the effects of weather removal \pm standard error.

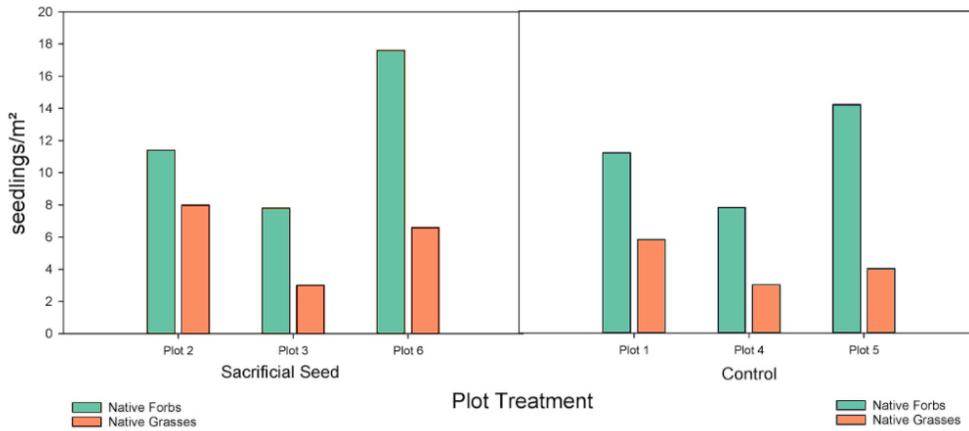


Figure 3 – Comparative seedling establishment of native forbs and native grasses between control plots and sacrificial seed plots. Twenty randomly selected points within a randomly selected 10 meter by 10 meter section of each plot were used to count emerging native forbs and grasses. Each point was sectioned using a 0.25m² metal frame as the border for counting. When comparing the control and sacrificial seed plots, it showed no significant difference. Data is shown as the number of emerging native forbs and grasses per m². Native grass data was generally lower than that of the forbs due to the nature of young grass seedlings being harder to identify and inexperience in identifying grasses.

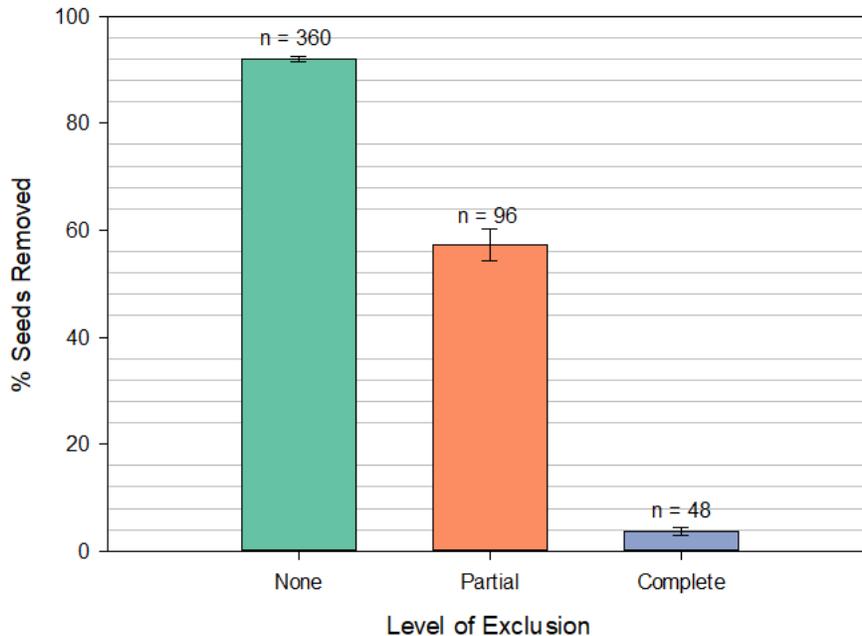


Figure 4 – Percentage of seed removal from seed cards with varying levels of predator exposure: exposed seed cards with no protection (none), seed cards from the original control cages which excluded small mammals, earthworms, and birds but not insects (partial), and the modified control cages which excluded all seed predators (complete) shows a rough trend in the predation levels from different granivores. Data is shown as mean percentage of seeds removed from seed cards with different exposure levels ± standard error. N has been included to show the large differences in the number of cards included at each exposure level. The data between exclusion levels also varied heavily in the time they were collected. Exposed card data came from the last three trials (6/12 – 7/16/2020), partial data came from trials four and five (6/12 – 7/2/2020), and the complete data came from trial six (7/9 – 7/16/2020).

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