Cheatgrass, Mammals, Birds, Butterflies, and Wildfire: A Study of Ecosystem Interactions

Running Head

Cheatgrass, Wildlife, and Wildfire

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AM, SJ, CM selected plot locations; AM, CM installed cameras and delineated plots; AM, SJ, CM conducted bird and butterfly counts; AM, CM conducted vegetation counts; AM, SJ analyzed images from cameras; AM, SJ, TS conducted analyses; AM, SJ drafted manuscript; AM, SJ, CM, TS reviewed and finalized manuscript.

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Executive Summary

Over two years of research, we used remotely triggering cameras, point counts, and transect counts to measure the number of mammals, birds, and butterflies across eight plots in the Rocky Mountain foothills that varied in cheatgrass cover. We found that mammals and butterflies were less numerous in areas infested with cheatgrass and that such areas sustained fewer bird and butterfly species. Furthermore, butterflies appeared more sensitive to small changes in cheatgrass cover than mammals. Cheatgrass density decreased dramatically on heavily infested plots burned by the Calwood fire (at low intensity), consistent with the findings of other research projects in similar ecoregions and climates. These areas regenerated with native plants and appeared to receive more use from wildlife than before the fire.

Management Implications

- Habitat restoration efforts and critical habitat designations should recognize that areas infested with cheatgrass hold lesser value to mammals and butterflies.

- Minor cheatgrass infestations may be of lesser concern in managing large mammals than butterflies, although major infestations may significantly degrade habitat for both taxa.

- Prescribed fire may be an effective tool for controlling cheatgrass under certain ecological and climactic conditions.

- Cheatgrass control methodologies should strive to restore holistic ecosystem function, including plant community composition and wildlife prevalence, instead of simply prioritizing proxies such as reductions in cheatgrass density or increased abundance of native species.

22   **Abstract**

23   Understanding the impacts of cheatgrass (*Bromus tectorum*), an invasive annual grass, on mam-

24   mal, bird, and butterfly populations is vital for wildlife habitat conservation, especially as cheat-

25   grass continues to spread across the western United States. The impact of cheatgrass on most

26   wildlife populations and their distributions has not been adequately studied. In this study, we in-

27   vestigate the impacts of cheatgrass on wildlife in the Rocky Mountain foothills in central Colo-

28   rado during 2020 and 2021. We used a combination of remote-triggering cameras, point counts,

29   and transect counts to quantify numbers of mammals, birds, and butterflies (respectively) on

30   eight 75m radius circular plots varying in cheatgrass cover. Cheatgrass cover was quantified dur-

31   ing each research season using a radial point-intercept method. Our findings indicate that both

32   mammals and butterflies avoid areas infested with cheatgrass. The extent of cheatgrass cover did

33   not appear to impact bird numbers or species richness of mammals significantly, but was a nega-

34   tive predictor of bird species richness and butterfly species richness. Our study suggests that

35   cheatgrass infestation can degrade habitats for mammals and butterflies and should be considered

36   when designing wildlife habitat conservation efforts. In late 2020 the Calwood fire burned part

37   of the research area, allowing us to investigate the impact of wildfire on cheatgrass. Our results

38   indicate that fire can significantly reduce cheatgrass cover within this ecosystem, and that pre-

39   scribed fire may be a potential tool for cheatgrass management under certain environmental con-

40   ditions.

41

44   **Introduction**

45   Cheatgrass (*Bromus tectorum*) spread rapidly through the western United States,

46   drastically altering native plant communities (Knapp 1996) and fire regimes (Bradley et al.

47   2018). Despite the environmental impacts of cheatgrass, few studies have focused on the role of

48   cheatgrass as an environmental stressor on wildlife populations. Assessments of habitat quality

49   in conservation and restoration projects should account for the prevalence of cheatgrass and its

50   impacts on native species. Furthermore, an improved understanding of cheatgrass-fire

51   interactions is important for developing sustainable approaches to mitigating negative impacts.

52   This study tested our hypothesis that cheatgrass infestation negatively impacts large

53   mammals, birds, and butterflies. We used remote-triggering cameras, point counts, and transect

54   counts to quantify the presence of these three types of animals on plots containing varying

55   amounts of cheatgrass. During the second research season in 2021, we tested an additional

56   hypothesis that wildfire affects cheatgrass density by comparing our 2020 and 2021 vegetation

57   data across both burned and unburned plots.

58   Ungulate diets primarily consist of native vegetation, although deer and elk consume

59   cheatgrass (Kohl et al. 2012). The movements of ungulates and other mammals likely reveal hab-

60   itat suitability more accurately than dietary studies, but we are unaware of studies investigating

61   how cheatgrass impacts large mammal distributions.

62   Among studies investigating the impact of cheatgrass on small mammals, dietary studies

63   produce results distinct from those of population studies. Even though Richardson et al. (2013)

64   found that cheatgrass seeds constituted the majority of seeds collected in cheek pockets of Great

65   Basin pocket mice (*Perognathus parvus*), numerous other studies have found significant de-

66   creases in mouse populations and diversity in areas infested by cheatgrass. One such study by

67   Ostoja & Schupp (2009) found that total rodent abundance in the Great Basin was 6.1 times

68   greater in sagebrush-dominated areas relative to cheatgrass-dominated areas. Similarly, Freed-

69   man et al. (2014) found that the abundance and diversity of small-mammal communities in the

70   Great Basin decreased with an increased abundance of cheatgrass.

71   These small mammal studies suggest that invasive plants that constitute a portion of ani-

72   mal diets are not necessarily beneficial to local animal populations. Therefore, we hypothesized

73   that large mammal densities would be lower in areas heavily infested by cheatgrass.

74   Birds appear to follow a similar pattern to mammals: dietary studies show that, although

75   cheatgrass seeds are palatable to some species, birds preferentially feed on native grasses (Goe-

76   bel & Berry, 1976). Under many of the environmental conditions found in the western U.S.,

77   Cheatgrass outcompetes native vegetation, possibly leading to a reduction in preferred food

78   sources for birds. Furthermore, arthropods, which constitute a significant portion of many bird

79   diets (Rotenberry, 1980), are affected by cheatgrass infestation (an interaction we discuss further

80   in the following paragraph). These observations lead us to suspect that cheatgrass infestation im-

81   pacts bird populations by altering food availability, forming our hypothesis that areas dominated

82   by cheatgrass would sustain decreased bird densities.

83   Little research has looked specifically at the interactions between cheatgrass and butter-

84   flies. However, extensive research on arthropod response to cheatgrass infestation shows a gen-

85   eral trend toward increased arthropod numbers in cheatgrass-infested areas, although this trend

86   does not hold for all taxa. Ostoja et al. (2009) and Gardner et al. (2009) found consistent in-

87   creases in arthropod numbers in cheatgrass-infested areas. However, Thapa-Magar et al. (2020)

88   found increased numbers of below-ground nesting native bees but reduced numbers of above-

89   ground nesting native bees in areas infested by cheatgrass in the Colorado Front Range. Pei et al.

90   (2023) found that litter accumulation from invasive grasses constrain native bee populations, alt-

91   hough they did not specifically investigate the impact of cheatgrass. A study conducted by

92   Looney & Zack (2008) found that beetle populations in cheatgrass-infested areas tended toward

93   herbivorous species.

94   Young et al. (1987) found that cheatgrass forms monocultures that outcompete other

95   plants, including potential nectar sources. Fleishman et al. (2005) found that butterfly numbers in

96   the Muddy River drainage (Nevada) were not impacted by non-native plants but were instead in-

97   fluenced by nectar availability. Based on these two studies that together suggest cheatgrass may

98   be detrimental to butterflies, we formed our hypothesis that cheatgrass-infested areas would sup-

99   port decreased butterfly numbers and butterfly species diversity.

100   Cheatgrass infestation generally increases fire risk and frequency (Bradley et al. 2018;

101   Whisenant, 1990), favoring cheatgrass regrowth over native vegetation (Melgoza & Nowak,

102   1991). Mitigating this positive feedback cycle has become a management priority to protect

103   intact ecosystems (Pilliod et al. 2021), with a particular focus on preserving ecosystems that are

104   maladapted to frequent fire regimes, such as the Great Basin sagebrush steppe. Despite the

105   significant environmental differences between the Great Basin sagebrush steppe and the Rocky

106   Mountain foothills, we hypothesized that we would observe a similar pattern of increased

107   cheatgrass abundance after fire.

108   **Methods**

109   **Study Area**

110   The study area lies at elevations between 1684-1790 m at the base of the Colorado Front

111   Range foothills in the United States, where the Great Plains converge with the Southern Rocky

112   Mountains. Annual precipitation averages 45-50 cm/year, with approximately half of this amount

113   falling during the March-June spring growing season (from US Climate Data 2020).

114   In early May 2020, we established eight circular 75 m radius plots on Boulder County

115   Parks and Open Space properties (Appendix A) near Lyons, Colorado (between 40°09'54.11" N,

116   105°16'16.54" W and 40°10'57.43" N, 105°15'28.98" W) where the Level IV ecoregion varies

117   from Foothills Shrubland to Front Range Fans (Chapman et al., 2006). During the first research

118   season, we located four plots in areas visually estimated to have high cheatgrass cover (> 20%)

119   and four plots in areas visually estimated to have low cheatgrass cover (< 20%). The visual esti-

120   mates were later confirmed by vegetation counts. We sampled the same plot locations in 2021.

121   Vegetation in the study area (scientific names follow Wittmann & Weber 2011, except

122   cheatgrass for which we use the widely accepted name, *Bromus tectorum*) consisted primarily of

123   foothills mixed-grass prairie, foothills shrub, and ponderosa pine woodland (Baker & Gala-

124   towitsch 1985; Colorado Natural Areas Program 1998). In relatively flat areas with deep soils,

125   these grasslands tend to be dominated by green needlegrass (*Nassella viridula*) and western

126   wheatgrass (*Pascopyrum smithii*), with numerous other native grass species well represented.

127   Flat-bottomed ravines support dense stands of native shrubs, including skunkbrush (*Rhus trilo-*

128   *bata*), wild plum (*Prunus americana*), and mountain mahogany (*Cercocarpus montanus*). Can-

129   yon bottoms support scattered hackberry trees (*Celtis reticulata*) and box elders (*Negundo ac-*

130   *eroides*) along with small patches of native tallgrasses, including switchgrass (*Panicum virga-*

131   *tum*) and indiangrass (*Sorghastrum avenaceum*). Ponderosa pines (*Pinus ponderosa*) are scat-

132   tered throughout the study area, becoming most numerous in rocky uplands. Non-native cheat-

133   grass is most prevalent in rocky or sloped areas.

134   All but one of the plots contain areas previously treated with the herbicides Esplanade

135   and Glyphosate, sometimes mixed with a combination of Quinstar, Dicamba, and/or Hardball

136   (Appendix A). The treatments occurred between 2017 and early 2020 prior to the first research

137   season. As we were not aware of these treatments until late in the research study, we did not fac-

138   tor the treatments into our experimental design or our choice of plot locations. Mensah et al.

139   (2015) identified glyphosate as a potentially significant toxin for wild mammal populations.

140   However, McComb et al. (2008) found that glyphosate exposure in wild animals remained well

141   below acute toxicity and did not appear to impact behavior. Van Deynze et al. (2022) found that

142   glyphosate application wasn't correlated with declines in butterfly numbers despite evidence for

143   genotoxicity to butterflies (Santovito et al. 2020). Given the limited impact of such treatments on

144   immediate animal behavior and fitness within treated areas, we expect that the prior herbicide

145   applications did not significantly impact our results.

146   A 4092 ha wildfire (Haverfield, 2021) on October 17th, 2020 (known as the "Calwood

147   fire") burned four of our plots (Appendix B), including two with high cheatgrass cover and two

148   with relatively low cheatgrass cover. In 2021, using the same methodology as in 2020, we re-

149   measured cheatgrass cover within burned and unburned plots and repeated the wildlife counts.

150   Although the Calwood fire burned intensely through much of its range, spectral reflectance anal-

151   ysis conducted by local agencies found that our research area experienced low soil burn severity

152   (*Cal-Wood Fire Rehab: Soil Burn Severity*, 2020). Our on-site observations, using burn severity

153   characteristics modified from Ryan and Noste (1985) by Neary et al. (2005), indicated that the

154   burn severity across the four burned plots was primarily "Light," apart from areas adjacent to

155   shrubs and trees that often experience "Medium" severity. The burned plots regenerated with na-

156   tive grasses and forbs, including dense patches of big bluestem (*Andropogon gerardii*) in some

157   previously cheatgrass-infested areas. The most intensely burned areas around fuel sources, such

158   as shrubs and bushes, regenerated with invasive thistles (*Carduus* spp.) and mustards (*Brassica*

159   spp. and *Sisymbrium* spp.), likely due to significant soil disturbance and diminishment of the na-

160   tive seed bank.

161   **Vegetation Sampling**

162   We conducted vegetation sampling using an identical methodology across both study

163   years. Within each plot, we established 40 sampling locations arranged in a radial pattern ad-

164   justed for the non-linear increase of circle area with increased circle radius (Figure 1). At each

165   sampling location, we used a Point-Intercept with a Grid Quadrant Method similar to that of

166   Caratti (2006), utilizing a 0.7-meter sampling frame with 25 sampling points arranged in a grid

167   pattern. We lowered a sampling pin at each sampling point and identified the vegetation beneath

168   the pin as cheatgrass plants, cheatgrass litter, bare ground and rocks, non-cheatgrass litter, or

169   non-cheatgrass plants.

170   **Mammal Sampling**

171   We mounted Stealth Cam (headquartered in Arlington, Texas, United States) G45NGX

172   remote-triggering cameras 0.9 meters off the ground on T-posts located at the center of each plot,

173   attached such that each camera could be oriented in any cardinal direction. The cameras were set

174   to maximum sensitivity with a 15-second recovery time to avoid missing mammals while keep-

175   ing false positive detections manageable.

176   We began observation at 2300 MST on May 13th and ended observation at 2259 MST on

177   August 13th in both 2020 and 2021. At intervals ranging from one to two weeks, we rotated the

178   cameras by 90 degrees, such that they completed two full rotations around their posts over the

179   course of the study.

180   We identified and counted all mammals in the images from the remote-triggering cam-

181 eras without relying on previous or subsequent images to detect or identify each mammal. Indi-

182 vidual mammals were counted regardless of whether they may have appeared in previous im-

183 ages. We categorized all detected mammals by species, or when not identifiable to the species, as

184 deer, ungulates, or mammals.

Bird Surveys

186   We counted birds seen or heard perching or foraging within 75m of the center points of

187 the eight monitoring plots for 8 minutes per plot on four mornings between 29 May and 10 July,

188 beginning each survey at sunrise and completing sampling of all eight plots by 0730 MST. We

189 varied the order of the plot counts during each replication to reduce temporal biases that might

190 stem from sampling bird populations at varying times of the early morning. As we entered each

191 plot, we counted any birds flushed from the vegetation. We counted swallows when they were

192 flying below the tops of the tallest trees and shrubs and when their irregular movement patterns

193 suggested that they were foraging. We did not count birds flying over or through the plots with-

194 out foraging (Ralph et al. 1998).

Butterfly Surveys

196   We counted butterflies seen along 150 m north/south transects bisecting each monitoring

197 plot during a time interval of 5 minutes per plot on six days in 2020 and four days in 2021 be-

198 tween 31 May and 10 August. We walked slowly (2 km/hr) along each transect, using binoculars

199 and cameras with telephoto lenses to identify all butterflies seen within 30 m. Counts were car-

200 ried out between 0745-0945 MST on calm (peak wind velocity $\leq$ 20 km/hr) and clear (mean

201 cloud cover $\leq$ 30%) mornings when the air temperature exceeded 18° C. We rotated the order of

202 plot sampling during each count replication to reduce temporal biases.

203   **Additional Data Collection**

204   In addition to measuring our primary variables, we quantified other properties of the

205   plots, notably slope, shrub and tree cover, and distance to a highway (effectively the distance to

206   the nearest human development). We also used vegetation sampling data to calculate the bare

207   and rocky ground percentage in each plot.

208   In 2020 we estimated woody plant (tree and shrub) cover using a custom grid overlay on

209   Google Earth Pro satellite imagery. We could not use the same methodology in 2021, as the

210   Google Earth imagery had not been updated since the Calwood fire. Instead, in 2022 we used a

211   different methodology to estimate woody plant cover, noting for each vegetation sampling

212   square whether it was located below a living tree or shrub or below a burned tree or shrub.

213   Shrubs that were burned to the ground or retained only a few charred stems were ignored.

214   Data from each of the two years were used to calculate both living and total (including

215   burned) woody plant cover, but shrub and tree density data were not compared across the two

216   years due to inconstancy in the data gathering methodologies.

217   **Data Analysis**

218   We used the vegetation sampling data to produce a cheatgrass cover value, representing

219   the percentage (expressed as a decimal) of each plot covered by either cheatgrass plants or cheat-

220   grass litter. We used cheatgrass cover as the independent variable in all primary calculations and

221   analyses, all of which were performed in the software package R.

222   We designated a Type 1 error rate of 5% ($p<.05$) as the statistical significance level in all

223   our analyses.

224   ANCOVA best fit our animal data as it allowed us to determine the relationship between

225   the number of animals and cheatgrass while adjusting for the sampling year. However, only our

226   2020 data consistently passed normality and homogeneity of variance tests (determined using the

227   Shapiro-Wilk Normality Test and the Fligner-Killeen Test of Homogeneity of Variances, respec-

228   tively). We used ranked animal data in the two-year animal-related analyses to accommodate the

229   different data normalizations required across the two years. We excluded interactions between

230   year and cheatgrass cover from the ANCOVA models because half the plots were burned in the

231   second year, thus significantly altering their cheatgrass cover relative to unburned plots.

232   We used R-squared values from Two-Way ANOVA and ANCOVA analyses to deter-

233   mine the best fit models for different animal categories to see if these values provide additional

234   insight into how different species categories respond to cheatgrass infestation. However, as de-

235   scribed earlier, we treated this purely as a secondary analysis and consistently used ANCOVA to

236   test our primary hypotheses.

237   One-Way ANOVA was used for secondary analyses comparing cheatgrass cover across

238   burn conditions. For these analyses, we used percent change in cheatgrass cover, calculated as

239   the change in cover across both years divided by the original cheatgrass cover of the plot. Data

240   assumptions were tested using the aforementioned normality and homogeneity of variance tests.

241   Data that failed these tests were normalized using Tukey Tests.

242   We additionally used Pearson's Correlation Coefficient Matrices to investigate other vari-

243   ables and interactions that may have influenced the results. We again used Tukey tests on the pri-

244   mary dependent variables to ensure normality. Ranking was not necessary because we could not

245   compare data across the two years using this method. We retained a Type I error rate of 5% due

246   to the limited number of variables.

247

248

249   **Results**

250   **Vegetation**

251   Cheatgrass cover on low cheatgrass plots varied from 0.031 to 0.135 in 2020, while

252   cheatgrass cover on high cheatgrass plots varied from 0.235 to 0.350 (Figure 2). The mean val-

253   ues of the two groups were statistically significant during the first year, $F(1,6)=44.72$, $p<0.001$,

254   confirming our original cheatgrass cover designations based on visual estimates.

255   We found a significantly higher percent decrease in cheatgrass cover, $F(1,6)=25.21$,

256      $p=.002$, and cheatgrass litter, $F(1,6)=57.95$, $p<.001$, on burned plots relative to unburned plots.

257   We also observed a greater percent decrease of cheatgrass plants in burned areas, but this effect

258   was not statistically significant, $F(1,6)=2.656$, $p=.15$. However, this effect was significant when

259   including only the four plots identified as high cheatgrass during the first research season,

260      $F(1,2)=334.7$, $p=.003$.

261   **Mammals**

262   During the total of 6 months (across two years) of remote camera observation, we rec-

263   orded 27,101 images, 752 (2.7%) of which contained at least one mammal. We detected 1091

264   individual mammals, 88% of which were ungulates.

265   ANCOVA analysis showed a significant inverse correlation between the cheatgrass cover

266   of plots and the number of observed mammals, $F(1,13)=7.432$, $p=.02$ (Figure 3). Year was not a

267   significant predictor of mammal detections, $F(1,13)=3.896$, $p=.07$. Mammal species richness was

268   significantly correlated with year, $F(1,13)=6.647$, $p=.02$, but not cheatgrass cover,

269      $F(1,13)=1.931$, $p=.19$ (Figure 3). We found that mammal numbers and mammal species richness

270   best fit a 2-way ANOVA model.

271

272   **Birds**

273   During the total of eight bird surveys (across two years), we observed 33 species across

274   the 8 plots. Obligate ground-nesters (Wickersham, 2017), which come into frequent direct con-

275   tact with grassy vegetation, comprised 40.9% of all birds observed.

276   ANCOVA analysis showed an insignificant negative relationship between mean birds per

277   plot and cheatgrass cover, $F(1,13)=3.719$, $p=.08$ (Figure 4), but a significant relationship with

278   year, $F(1,13)=11.83$, $p=.004$. We found a significant negative relationship between bird species

279   richness and cheatgrass cover, $F(1,13)=6.8154$, $p=.02$ (Figure 4), along with a significant corre-

280   lation between bird species richness and year, $F(1,13)=16.39$ $p=.001$. Bird numbers best fit a 2-

281   way ANOVA model, while bird species richness followed an ANCOVA pattern.

282   **Butterflies**

283   During the total of ten butterfly surveys (across two years), we observed 33 butterfly spe-

284   cies. Variegated fritillaries, habitat generalists that occasionally invade the Rocky Mountain re-

285   gion in large numbers in response to environmental stresses in the southern United States (Opler

286   1999), comprised 69% of all butterflies observed.

287   ANCOVA analysis showed a significant negative relationship between mean butterflies

288   and cheatgrass cover, $F(1,13)=16.81$, $p=.001$ (Figure 5), but no correlation between mean butter-

289   flies and year, $F(1,13)=2.274$, $p=.16$. Butterfly species richness was significantly negatively cor-

290   related with cheatgrass cover, $F(1,13)=23.53$, $p<.001$ (Figure 5), and significantly correlated with

291   year, $F(1,13)=14.65$, $p=.002$. Both butterfly numbers and butterfly species richness best fit an

292   ANCOVA model.

293

294

295   **Additional Factors**

296   Analysis investigating relationships between the primary variables and plot slope, bare

297   ground, shrub and tree cover, and distance to human development generally supported our hy-

298   pothesis that cheatgrass cover was the primary driving factor influencing wildlife abundance (Ta-

299   ble 1; Table 2). However, mammals were negatively correlated with plot distance to human de-

300   velopment in 2021. Butterfly numbers in 2020 correlated negatively with plot distance to human

301   development, and butterfly species richness in 2021 correlated negatively with both plot distance

302   to human development and plot slope. We also found positive correlations between living woody

303   plant cover and cheatgrass cover, cheatgrass plants, and cheatgrass litter in 2021.

304   **Discussion**

305   **Confirmation of Primary Hypothesis**

306   Our findings support the hypothesis that cheatgrass negatively impacts habitat suitability

307   for native wildlife.

308   The two-year study indicates that large mammals, particularly ungulates, avoid areas in-

309   fested by cheatgrass. Given the predominance of ungulates in our dataset, we do not expect the

310   mammal results to represent a general mammal response to cheatgrass accurately. Ungulates are

311   probably sensitive to cheatgrass infestation due to their preference for grazing on grasses and

312   forbs that directly compete with cheatgrass. Other mammal species, particularly predators, are

313   unlikely to be similarly impacted by cheatgrass, which may explain the weak correlation between

314   mammal species richness and cheatgrass cover in our results.

315   Our study suggests a possible negative impact of cheatgrass infestation on bird numbers.

316   Bird density was generally lower on cheatgrass-infested plots, but this tendency was not statisti-

317   cally significant. However, bird species richness was significantly lower on cheatgrass-infested

318   plots. We examined the possibility that this inconsistency in observed bird response was due to

319   variation in shrub cover across the plots, but our data did not support this hypothesis.

320   Our study indicates that butterflies are strongly impacted by cheatgrass infestation. Both

321   butterfly abundance and species richness were significantly lower in areas infested by cheatgrass.

322   This is likely due to decreased nectar and host plant availability as cheatgrass displaces native

323   plants.

324   **Varied Species Group Responses to Cheatgrass**

325   The informal comparison of Two-Way ANOVA and ANCOVA models suggests that

326   mammals respond to discrete cheatgrass densities, while any increase in cheatgrass density im-

327   pacts butterflies. Due to their larger home ranges relative to butterflies, we hypothesize that

328   mammals are relatively unaffected by cheatgrass interspersed among other plants up to a certain

329   threshold level above which foraging in a particular area becomes unattractive. Conversely, but-

330   terflies have small home ranges and are dependent on native grasses and forbs as host plants and

331   nectar sources within a small area. Therefore, any reduction in native plant density, even within a

332   relatively small area, may directly drive down butterfly populations. Further research with larger

333   datasets and formal statistical tests could strengthen our tentative conclusion.

334   **Alternative Explanations**

335   Factors other than cheatgrass cover are unlikely to explain our results.

336   Although mammal numbers, butterfly numbers, and butterfly species showed a weak

337   negative correlation with distance to human development and plot slope, this was likely due to

338   high cheatgrass density plots being located farther away from the highway at higher elevations

339   characterized by steep and rocky slopes.

340   The correlation between living woody plant cover and cheatgrass is likely due to the par-

341   allel die-off of shrubs and reduction of cheatgrass density on burned plots.

342   **Impact of Fire on Cheatgrass**

343   Contrary to our original hypothesis, fire reduced cheatgrass infestation and favored native

344   plant recovery.

345   In the literature, cheatgrass is generally described as increasing fire risk and fire fre-

346   quency (Bradley et al. 2018; Whisenant, 1990). However, most cheatgrass studies have been

347   conducted in a limited set of locations (typically in the Great Basin area) that are not representa-

348   tive of all ecoregions (Porensky & Blumenthal, 2016). The literature suggests multiple key fac-

349   tors that could explain the difference between cheatgrass response to fire on our plots from the

350   response frequently observed in the Great Basin area: increased altitude, increased soil moisture,

351   decreased temperature, and remnant native seed bank. Urza et al. (2019) found increased re-

352   sistance to cheatgrass invasion after fire on higher elevation plots, which were consequently

353   cooler and moister, compared to lower elevation plots. Consistent with these findings, Sherrill &

354   Romme (2012) identified increased altitude, increased fire severity, and increased post-fire soil

355   moisture as factors decreasing cheatgrass return after fire. White and Currie (1983) found that

356   fall burns in Montana were more effective at controlling cheatgrass than spring burns, although

357   these researchers consistently observed decreases in cheatgrass density regardless of burn timing.

358   Lastly, evidence that reseeding of native vegetation after fire is effective at controlling invasive

359   species, including cheatgrass, over both short (Thompson et al. 2006) and long (Ott et al. 2019)

360   time periods suggests that a remnant native seed bank in infested areas would help reduce cheat-

361   grass regrowth post-fire.

362   Our plots satisfied many of the requirements for increased resistance to cheatgrass infes-

363   tation after fire, as they were located in a mid-elevation environment characterized by relatively

364   high winter seasonal precipitation (which occurred shortly after the fire) and relatively low tem-

365   peratures. Our plots also sustained remnant native vegetation in cheatgrass-infested areas, likely

366   indicating the presence of a dormant native seed bank. Furthermore, the Calwood Fire occurred

367   on October 17th, 2020, during the season that White and Currie (1983) identified as a particularly

368   effective season for fire-based cheatgrass control.

369   Our results, combined with previous studies, suggest that controlled fire may be an effec-

370   tive tool for cheatgrass control in Boulder County and possibly across the Colorado Front Range

371   and beyond. Because the Rocky Mountain foothills ecosystem is highly fire-adapted (Kaufmann

372   et al. 2006), the cheatgrass fire cycle observed in fire-sensitive Great Basin landscapes may not

373   be present in our area.

374   **Further Research**

375   Although our research provides insight into the impact of cheatgrass on large mammals,

376   birds, and butterflies, the impact of cheatgrass on wildlife populations requires further study to

377   inform sustainable management practices.

378   Our results should be verified and expanded through research covering a larger geo-

379   graphic area utilizing more plots, broader animal sampling, and extending over a longer time-

380   scale. Of particular interest would be the inclusion of multiple invertebrate taxa in sampling

381   methodology to understand better their contribution to the impact of cheatgrass on higher trophic

382   levels.

383   A variety of methods have been used to manage cheatgrass infestation. Future research

384   should investigate the differential impact of such methods on wildlife populations. We

385   recommend that such research compare wildlife populations over appropriately long timescales

386   on untreated plots relatively free of cheatgrass, on untreated plots infested by cheatgrass, and on

387   previously infested treated plots. The standard for treatment success should include restoration to

388   ecosystem conditions similar to those found in untreated, relatively cheatgrass-free areas, not just

389   improvement over untreated cheatgrass-infested areas. Failing to investigate the effect of cheat-

390   grass treatment methods on wildlife could potentially cause the inadvertent degradation of valua-

391   ble wildlife habitat. For example, some herbicides are known to negatively impact certain butter-

392   fly species (Russell & Schultz 2010). Without studies investigating the effects of herbicides on

393   butterflies, herbicides might be used when alternative methods, such as those suggested by Blu-

394   menthal et al. (2010), would be more appropriate.

395   Our results, combined with the literature on fire and cheatgrass interactions, suggest that

396   controlled fire may be a powerful cheatgrass management method in certain ecosystems and re-

397   gions. Further research should more precisely identify ecological and geographic limits within

398   which fire is a viable cheatgrass control method. Understanding the precise influence of both

399   pre- and post-burn precipitation on cheatgrass regrowth would aid managers in choosing the

400   ideal timing for prescribed burns aimed at controlling cheatgrass. Long-term monitoring and re-

401   search after both controlled and uncontrolled fires would deepen understanding of cheatgrass-fire

402   interactions and reveal the effectiveness of management strategies.

403   **Conclusion**

404   Our study indicates that mammals and butterflies are impacted by cheatgrass infestation, alt-

405   hough in different ways. Mammals respond to a threshold level of cheatgrass cover above which

406   the infested areas are undesirable for foraging. Conversely, butterflies respond to even small

407   changes in cheatgrass cover, likely due to their dependency on native plant availability within a

408   small area. While bird species richness is impacted by cheatgrass cover, we could not conclude

409   that bird numbers are similarly affected (possibly due to variations in other characteristics across

410   our plots). Future research focusing on large mammal, small mammal, bird, and invertebrate

411   populations could help refine the understanding of the impact of cheatgrass infestation on wild-

412   life.

413   Results following unexpected fire disturbance during our research indicate that fire can

414   reduce cheatgrass infestation in the Colorado Front Range foothills grassland ecosystem, sug-

415   gesting that prescribed fire is a tool for land managers addressing the environmental degradation

416   caused by this invasive annual grass.

417   **Acknowledgments**

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554

555   **Illustrations**

556   **Tables**

557   *Table 1. Table of p-values correlating each primary variable with additional factors in 2020,*

558   *specifically ranked number of mammals (Mam), ranked number of mammals species (Mam S),*

559   *ranked mean number of birds (Bird), ranked number of bird species (Bird S), ranked mean num-*

560   *ber of butterflies (Bfly), ranked number of butterfly species (Bfly S), plot Bare Ground Cover*

561   *(Bare G), plot distance to human development (DtH), and slope (Slope).*

	Slope	DtH	Bare G	Bfly S	Bfly	Bird S	Bird	Mam S
Mam	0.126	0.094	0.565	0.053	0.181	0.241	0.091	0.313
Mam S	0.179	0.055	0.612	0.114	0.104	0.068	0.218	
Bird	0.681	0.699	0.715	0.104	0.521	0.081		
Bird S	0.456	0.220	0.331	**0.014**	0.056			
Bfly	0.115	**0.041**	0.054	**0.014**				
Bfly S	0.227	0.180	0.277					
Bare G	**0.038**	0.066						
DtH	**0.005**							

562

563 *Table 2. Table of p-values correlating each primary variable with additional factors in 2021,*

564 *specifically ranked number of mammals (Mam), ranked number of mammals species (Mam S),*

565 *ranked mean number of birds (Bird), ranked number of bird species (Bird S), ranked mean num-*

566 *ber of butterflies (Bfly), ranked number of butterfly species (Bfly S), plot Bare Ground Cover*

567 *(Bare G), plot distance to human development (DtH), and slope (Slope).*

|	Slope	DtH	Bare G	Bfly S	Bfly	Bird S	Bird	Mam S
Mam	0.141	**0.037**	0.438	0.120	**0.043**	0.469	0.943	0.488
Mam S	0.198	0.156	0.598	0.117	0.895	0.415	0.491	
Bird	0.833	0.738	0.499	0.347	0.415	0.131		
Bird S	0.096	0.087	0.239	**0.024**	**0.049**			
Bfly	0.152	0.119	0.432	**0.030**				
Bfly S	**0.005**	**0.016**	0.153					
Bare G	**0.008**	0.078						
DtH	**0.005**							

568

569   **Figures**

570   *Figure 1. Diagram illustrating our modified radial distribution of plot sampling locations that*

571   *increases sampling uniformity for circular plots. Red points indicate sampling locations that de-*

572   *viate significantly from traditional radial sampling methods.*

573
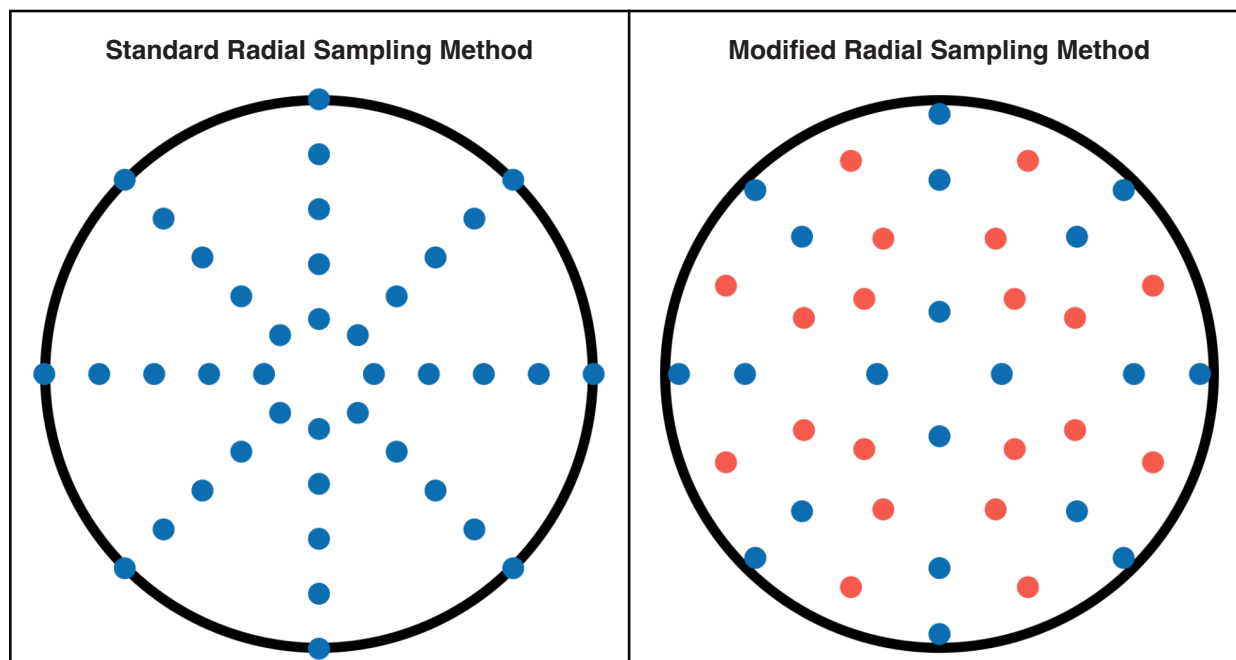
574   *Figure 2. Graph comparing cheatgrass density on each plot between 2020 and 2021 using stand-*

575   *ard boxplot notation. Blue data points indicate unburned plots, while red data points indicate*

576   *plots burned between 2020 and 2021. The data points are shifted horizontally to enhance the dif-*

577   *ference between the box plot and individual data points.*

578
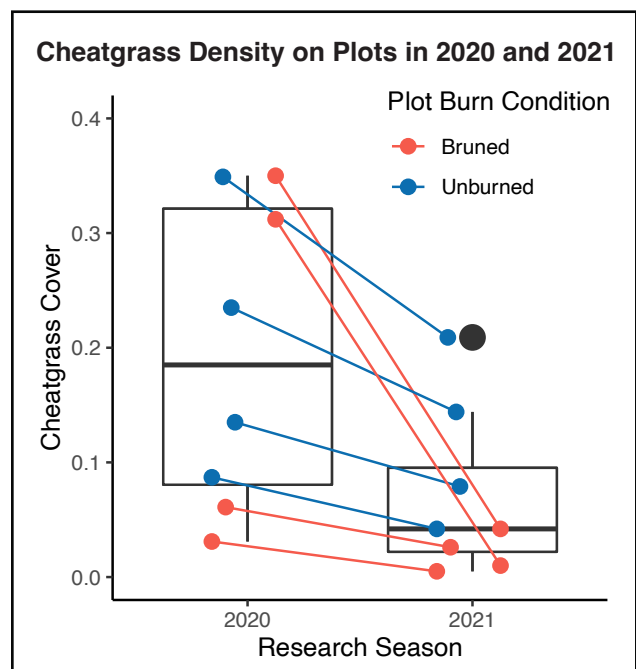
579   *Figure 3. Graph showing the ranked number of observed mammals and mammal species corre-*

580   *lated with cheatgrass cover on each plot for 2020 and 2021.*

581

582
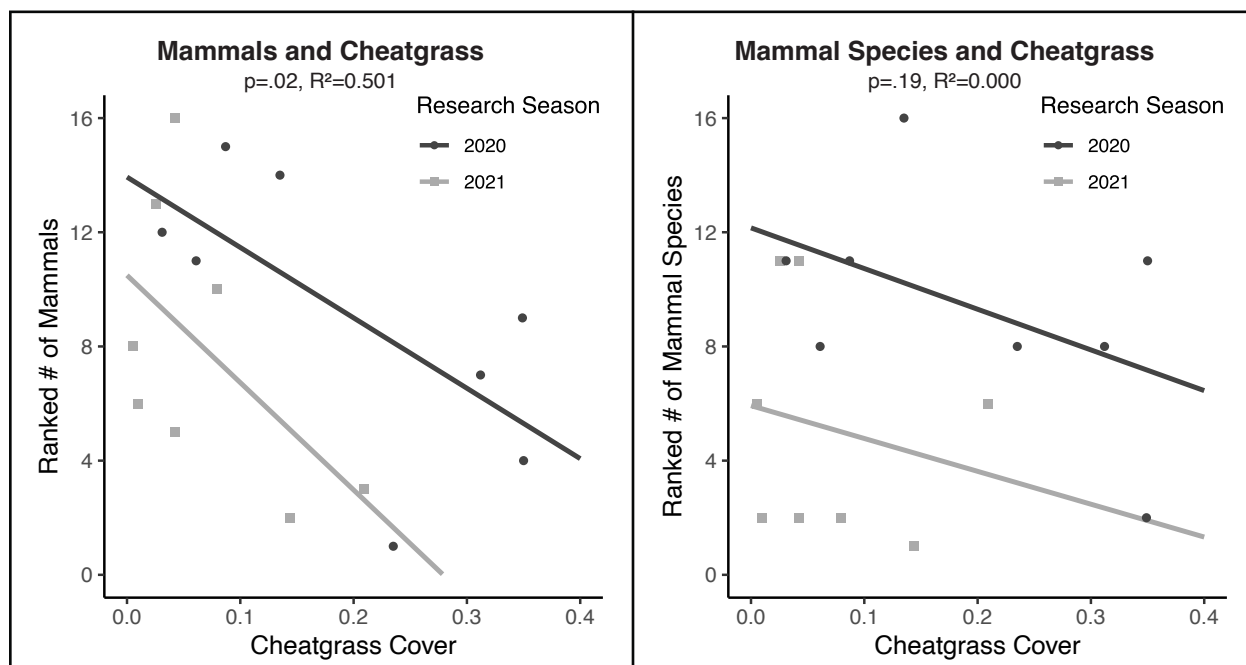
583   *Figure 4. Graph showing the ranked mean number of observed birds and bird species correlated*

584   *with cheatgrass cover on each plot for 2020 and 2021.*

585

586
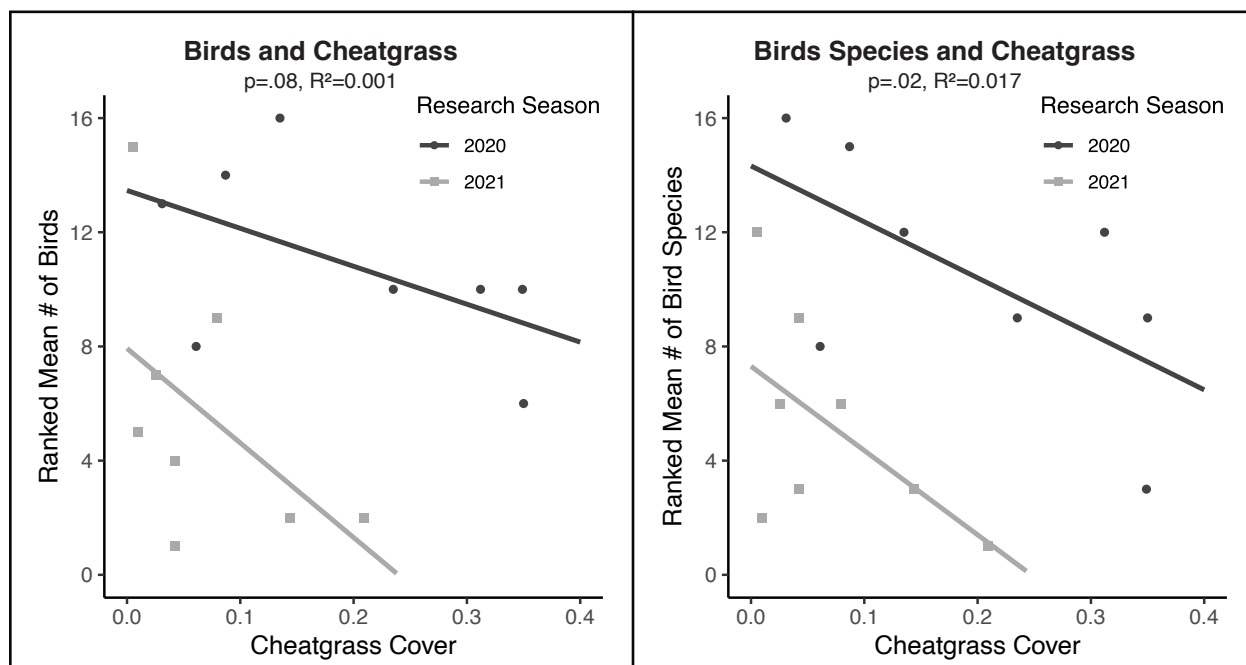
587   *Figure 5. Graph showing the ranked mean number of observed butterflies and butterfly species*

588   *correlated with cheatgrass cover on each plot for 2020 and 2021.*

589