Experimental removal of introduced slider turtles offers new insight into competition with a native, threatened turtle

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The red-eared slider turtle (*Trachemys scripta elegans*; RES) is often considered one of the world's most invasive species. Results from laboratory and mesocosm experiments suggest that introduced RES outcompete native turtles for key ecological resources, but such experiments can overestimate the strength of competition. We report on the first field experiment with a wild turtle community, involving introduced RES and a declining native species of conservation concern, the western pond turtle (*Emys marmorata*; WPT). Using a before/after experimental design, we show that after removing most of an introduced RES population, the remaining RES dramatically shifted their spatial basking distribution in a manner consistent with strong intraspecific competition. WPT also altered their spatial basking distribution after the RES removal, but in ways inconsistent with strong interspecific competition. However, we documented reduced levels of WPT basking post-removal, which may reflect a behavioral shift attributable to the lower density of the turtle community. WPT body condition also increased after we removed RES, consistent with either indirect or direct competition between WPT and RES and providing the first evidence that RES can compete with a native turtle in the wild. We conclude that the negative impacts on WPT basking by RES in natural contexts are more limited than suggested by experiments with captive turtles, although wild WPT do appear to compete for food with introduced RES. Our results highlight the importance of manipulative field experiments when studying biological invasions, and the potential value of RES removal as a management strategy for WPT.

1   **Full Title**: Experimental removal of introduced slider turtles offers new insight into competition

2   with a native, threatened turtle

4   **Short Title**: Turtle competition

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34

35   **Abstract**: The red-eared slider turtle (*Trachemys scripta elegans*; RES) is often considered one

36   of the world's most invasive species. Results from laboratory and mesocosm experiments

37   suggest that introduced RES outcompete native turtles for key ecological resources, but such

38 experiments can overestimate the strength of competition. We report on the first field experiment

39 with a wild turtle community, involving introduced RES and a declining native species of

40 conservation concern, the western pond turtle (*Emys marmorata*; WPT). Using a before/after

41 experimental design, we show that after removing most of an introduced RES population, the

42 remaining RES dramatically shifted their spatial basking distribution in a manner consistent with

43 strong intraspecific competition. WPT also altered their spatial basking distribution after the RES

44 removal, but in ways inconsistent with strong interspecific competition. However, we

45 documented reduced levels of WPT basking post-removal, which may reflect a behavioral shift

46 attributable to the lower density of the turtle community. WPT body condition also increased

47 after we removed RES, consistent with either indirect or direct competition between WPT and

48 RES and providing the first evidence that RES can compete with a native turtle in the wild. We

49 conclude that the negative impacts on WPT basking by RES in natural contexts are more limited

50 than suggested by experiments with captive turtles, although wild WPT do appear to compete for

51 food with introduced RES. Our results highlight the importance of manipulative field

52 experiments when studying biological invasions, and the potential value of RES removal as a

53 management strategy for WPT.

54 **Introduction**

55 The International Union for Conservation of Nature (IUCN) has labeled the red-eared slider

56 turtle (*Trachemys scripta elegans*; RES) one of the "world's worst invasive species" (Lowe et al.

57 2000). RES are native to the central United States —from West Virginia through Texas to

58 eastern New Mexico and north into Illinois—but are now established throughout much of the

59 country. RES are also globally distributed, having been released on every continent except

60 Antarctica with robust populations in Europe and Asia. These introductions predominantly result

61 from the release of unwanted pet turtles (Kraus 2009; Rhodin et al. 2017). Results from

62 laboratory and mesocosm experiments suggest that RES can outcompete native European and

63 eastern North American freshwater turtles for food and basking sites (Cadi and Joly 2003, 2004;

64 Polo-Cavia et al. 2010, 2011; Pearson et al. 2015). While such controlled experiments are

65 informative, they can also inflate the effects of competition compared to *in situ* field

66 manipulations (Skelly 2002; Winkler and Van Buskirk 2012). Comparing laboratory and

67 mesocosm experiments with field manipulations is a critical step to a more complete

68 understanding of the strength and mechanisms underlying species interactions in nature.

69 However, to our knowledge, no study has experimentally tested for competition between non-

70 native RES and any native turtle species in the wild.

71 Basking sites are a key resource for thermoregulation, disease control, and reproduction

72 in freshwater turtles (Ernst and Lovich 2009), and previous *ex situ* experiments suggest that

73 basking sites are an important axis of competition between native turtles and introduced RES

74 (Cadi and Joly 2003; Polo-Cavia et al. 2010). Prior work in the University of California, Davis

75 Arboretum waterway (hereafter, UCD Arboretum) found that introduced RES and native western

76 pond turtles (*Emys marmorata*; WPT) sometimes bask at the same sites (Fig 1), although they

77 tend to use basking sites that differ physically and spatially (Lambert et al. 2013). In particular,

78 WPT and RES predominantly bask in the western and eastern ends of the UCD Arboretum,

79 respectively. While WPT basking is not related to particular habitat characteristics, RES basking

80 is related to sites with more human activity, steel mesh basking substrates, deeper water, and

81 shallower slopes. Whether these basking site differences are the result of species-specific habitat

82 choices or competition has never been resolved and requires an experimental approach.

83   Many freshwater turtles, including both WPT and RES, are dietary generalists as adults

84   and consume a broad array of food items, though they tend to shift from higher rates of carnivory

85   when young to higher rates of herbivory as adults (Ernst and Lovich 2009). Even so, laboratory

86   and mesocosm experiments suggest RES might directly interfere with native turtle food

87   consumption through aggressive behaviors or higher food consumption rates, which can limit

88   food availability and growth rates of less competitive turtles (Cadi and Joly 2004; Polo-Cavia et

89   al. 2011; Pearson et al. 2015). Additionally, if turtle densities are high for a given habitat,

90   exploitative competition could limit food availability, both intra- and interspecifically, and

91   therefore decrease growth rates and / or body condition of native species.

92   Here, we present the results of an *in situ* field experiment where we substantially reduced

93   the introduced RES population at the UCD Arboretum to test for competition with WPT.

94   Because the waterway is disjunct from other turtle populations, the UCD Arboretum represents a

95   closed system for WPT that is well suited for experimental manipulations; natural

96   immigration/emigration is not possible for freshwater turtles in this system, although occasional

97   human-assisted transport does occur, particularly with RES released into the waterway. Our

98   experiment is the first to explicitly test whether invasive species removal, a commonly-

99   advocated management practice for invasive species including RES (Gaeta et al. 2015; García-

100   Díaz et al. 2017), influences the basking behavior and body condition of a native turtle in the

101   wild. If the distribution of WPT basking is a result of direct, competitive exclusion by RES from

102   optimal basking sites, then RES removal should result in an increase of post-removal WPT

103   basking at sites previously dominated by RES. Alternatively, if WPT basking activity does not

104   significantly change in this manner after RES removal, then existing behavioral basking

105   differences between the two species likely reflect species-specific habitat preferences,

106   competitive superiority of WPT, or both. We also assessed WPT body condition pre- and post-

107   removal as a proxy for whether removing RES improves WPT access to food resources. If

108   introduced RES compete with WPT for food, then removing RES should result in an increase in

109   WPT body condition. Given the broad overlap of these two species across California (Thomson

110   et al. 2010, 2016), the range-wide imperilment of WPT (Spinks et al. 2003; Thomson et al.

111   2016), and the current Status Review for possible WPT listing under the U.S. Endangered

112   Species Act (USFWS 2015), this experiment is directly relevant to ongoing WPT management

113   actions.

114

115   **Methods**

116   UC Davis IACUC Protocols #15263 and #16227 and California Department of Fish and Wildlife

117   Scientific Collecting Permits #2480, #4307, and #11663 approved this work. We conducted all

118   analyses in the R statistical language (version 3.5.2).

119

120   *Study Site*

121   Our study took place at the UCD Arboretum (38.53, -121.76), a permanent waterway extending

122   along the southern border of the UC Davis campus, Yolo County, California, USA (Fig 1). The

123   UCD Arboretum was formed in the 1870s when the historical north fork channel of Putah Creek

124   was diverted into the south fork (Larkey 1980). This waterway is 2.4 km long, ca. 4 ha in surface

125   area, and averages 15 m wide and 1 m deep (Spinks et al. 2003). Terrestrial habitat surrounding

126   the waterway is irrigated and landscaped with predominantly non-native vegetation (Spinks et al.

127   2003). A 1.5–2.5 m wide paved path encircles the entire waterway within 5–10 m of the water's

128   edge. This path is regularly used by pedestrians, bicyclists, and maintenance vehicles which

129   influence turtle basking (Lambert et al. 2013, Costa 2014). The waterway's shoreline—including

130   basking sites— is a combination of concrete, exposed dirt, and landscaping steel mesh which has

131   been exposed by erosion (Lambert et al. 2013).

132

133   *Turtle trapping and RES removal*

134   Across the UCD Arboretum, we deployed baited submersible traps in optimal habitat for both

135   RES and WPT over approximately 900 trap-days from 10 July–1 August, 2011 and again from

136   13–29 September, 2011. We supplemented this trapping with dip netting and opportunistic hand

137   captures during both periods, and with a fyke net and a basking trap during the latter period. Dip

138   netting and hand captures were targeted at RES but other trapping was not. We removed and

139   euthanized all RES, depositing most specimens at the UC Davis School of Veterinary Medicine,

140   the Natural History Museum of Los Angeles County, or the UC Davis Museum of Wildlife and

141   Fish Biology. Beginning in the 1996, our group uniquely marked each captured WPT with scute

142   notches using a handheld file or (juveniles only) nail clippers (Spinks et al. 2003). We similarly

143   marked any new WPT during this trapping effort. We used linear regression to test whether our

144   trapping depleted the RES population over time by regressing cumulative RES captures against

145   trapping day for adult RES (Krebs 1989). Using likelihood ratio tests (Crawley 2013) to assess

146   model fits, we compared a quadratic model, which would indicate population depletion, to a

147   linear model, which would indicate that the RES population was not leveling off with our

148   removal effort.

149

150   *WPT Body Condition*

151 To estimate changes in WPT body condition, we trapped for one week the year following RES

152 removal, from 27 May–2 June, 2012. Due to logistical constraints we were unable to trap later in

153 the summer at a similar time as in 2011. Differences in trapping dates may influence body

154 condition analyses because females may be gravid and therefore heavier in the earlier 2012

155 sampling or because all turtles may have had more time to put on mass during the later 2011

156 sampling. However, these effects are likely limited. In both 2011 and 2012 we measured WPT

157 plastron length (notch-to-notch; mm) with dial calipers and body mass (g) with Ohaus CS2000

158 digital pan scales (Iverson and Lewis 2018). We used a linear mixed-effects model (function

159 'lmer', R package "lme4"; Bates et al. 2015) to test whether WPT body condition (i.e.,

160 differences in mass controlling for body length) changed after the RES removal (Cadi and Joly

161 2003; Schulte-Hostedde et al. 2005; Litzgus et al. 2008). Our model of WPT mass controlled for

162 plastron length and included treatment (pre- or post-removal) and sex as fixed effects and

163 individual WPT as a random effect to control for repeated measures. This model simultaneously

164 regresses body mass against plastron length and tests for differences in the residuals of this

165 model between study year and sex. We used likelihood ratio tests to assess the significance ($\alpha <$

166 0.05) of fixed effects and removed non-significant variables from our model. We obtained full

167 model conditional R^2 (cR^2) for fixed and random effects combined and a marginal R^2 (mR^2) for

168 the model's fixed effects alone (function 'r.squaredGLMM', package "MuMIn", Barton 2018).

169

170 *Basking Site Monitoring*

171 We conducted binocular surveys of 24 pre-selected basking sites (Fig 1) for 34 total days—16

172 days pre-removal between 18 March and 22 April 2010 (Lambert et al. 2013) and 18 days post-

173 removal between 18 March and 22 April 2012. Following Lambert et al. (2013), we performed

174   all surveys between 1000 and 1500 hr to coincide with the expected maximum turtle basking

175   activity during this time of year. We surveyed all sites once daily in rapid succession to avoid

176   counting the same turtle at multiple sites. Each survey was performed in under one hour, at a

177   distance of ca. 10–100 m from turtles, and did not noticeably disturb basking turtles. MRL and S.

178   Nielsen conducted basking surveys in 2011 and JMM and RMS conducted 2012 surveys; all

179   surveyors were trained by GBP and HBS. During each survey we recorded the number of

180   individuals of each species basking at each basking site as well as water temperature because we

181   previously found that basking activity of both species increases more with warmer water

182   temperatures than with air temperatures (Lambert et al. 2013). We also obtained air temperature

183   data from the UC Davis Russell Ranch Weather Station which is located ca. 4 km NW of the

184   UCD Arboretum.

185

186   *Modeling the Effects of RES Removal on Turtle Basking*

187   We tested for changes in the relative basking distribution of WPT and RES (i.e., the ratio of

188   WPT:RES) across the UCD Arboretum pre- and post-RES removal using a generalized linear

189   mixed effects model (GLMM) with a binomial family for proportion data (function 'glmer', R

190   package "lme4"). A binomial GLMM accounts for binary data (two species here) and variation

191   in sample sizes across basking sites and survey days. We modeled WPT:RES basking as a

192   function of treatment (pre- or post-removal) and the distance of each basking site from the west

193   end of the UCD Arboretum because turtle basking distributions were previously shown to vary

194   west-east (Lambert et al. 2013). We accounted for repeated measures by treating survey date as a

195   random effect (Lambert et al. 2013). To explore site-specific changes in the ratio of the two

196   species, we also used individual binomial GLMMs for each basking site.

197   In addition, we modeled the absolute basking abundance of both species pre- and post-

198   removal using Poisson GLMMs (function 'glmer', R package "lme4") for count data. Our

199   approach here was the same as with the binomial GLMM and, if an interaction between

200   treatment and distance from the west end was significant, we used individual GLMMs for each

201   year to test the pattern and strength of turtle basking distributions across the UCD Arboretum in

202   each year. To test whether certain basking sites made up larger or smaller proportions of total

203   WPT basking observations pre- or post-removal, we used contingency tables, focusing on the

204   five most heavily-used turtle basking sites (combined for both species) pre-removal (sites P, O,

205   E, Q, and R) and site X, the most heavily-used turtle basking site post-removal.

206

207   **Results**:

208   *Trapping and RES Removal*

209   We removed and euthanized 177 RES (100.6 kg total biomass), including 28 adult males (16.3

210   kg), 72 adult females (79.4 kg), and 77 juveniles (4.9 kg, defined as $\leq$ 100 mm carapace length;

211   Ernst and Lovich 2009). A quadratic (rather than linear) model fit our data best (likelihood ratio

212   test $p < 0.0001$, full model $R^2 = 0.95$) and showed RES captures leveling off, signifying we had

213   removed a substantial fraction of the RES population.

214   We also captured, marked, and released 115 unique WPT (62.7 kg total biomass)

215   comprising 51 males (36.1 kg), 36 females (24.1 kg), and 28 juveniles (2.5 kg, defined as $\leq$ 110

216   mm plastron length; Holland, 1991).

217

218   *WPT Body Condition*

219   While we trapped a larger number of WPT in each year, we trapped 25 unique adult WPT in

220   both 2011 and 2012; we used these 25 WPT for the body condition analysis. The body condition

221   linear model showed no interaction between treatment and sex (p = 0.92) and so we removed this

222   interaction from the model. Sex (p = 0.009), treatment (p < 0.001), and plastron length (p <

223   0.0001) were significant (full model cR^2 = 0.95, mR^2 = 0.86). For a given plastron length, males

224   were on average 61.54 g (± 23.55 g) heavier than females. Although the 25 WPT measured

225   before and after RES removal showed individual variation in their degree of body condition

226   change post-removal (Fig 2A), on average they were 39.80 g (± 9.92 g) heavier for a given

227   plastron length post-removal (Fig 2B).

228

229   *Basking Site Monitoring*

230   We recorded 283 WPT and 645 RES observations in 2010 but only 43 WPT and 61 RES

231   observations in 2012. Although the reduction in numbers of observed WPT was unexpected, we

232   do not believe this reflects a decline in the WPT population. From 27 May–2 June 2012, we

233   trapped 54 unique WPT over seven days and a Schnabel multiple capture-mark-recapture

234   population estimate (Krebs 1989) derived from trapping data (beginning in the mid 2000s)

235   suggest that ca. 162 WPT (including 10 newly-marked juveniles) were present in the UCD

236   Arboretum immediately after our post-removal surveys (McKenzie, Screen, and Pauly unpubl.);

237   this estimate is similar to pre-removal estimates of WPT population size (ca.146 WPT, including

238   18 newly-marked juveniles). Given these estimates, we are confident that the WPT population

239   was essentially unchanged during our experiment, and thus our focus on the relative basking

240   distributions of turtles at monitored basking sites meaningfully reflects the impact of our removal

241   experiment and not a catastrophic decline in WPT.

242   The basking sites most commonly used by WPT pre-removal were generally the same

243   sites used post-removal (Fig 3A, B). We recorded WPT basking at 15 of 24 basking sites pre-

244   removal, but at only 8 of 24 sites post-removal. WPT were absent from 8 sites they used pre-

245   removal (although of these, only 2 were frequently used pre-removal: sites A and N) and were

246   present at one additional site where they were not recorded pre-removal (site B). We recorded

247   RES basking at 17 of 24 basking sites pre-removal, and only 8 of 24 sites post-removal (Fig 3A,

248   B). RES were absent from 9 sites they used pre-removal and were not recorded using new sites

249   post-removal.

250   Water temperatures were warmer in 2010 (17.0 C ± 0.24 SE) than in 2012 (15.4 C ± 0.36

251   SE; two-tailed t-test, p < 0.0001). However, maximum daily air temperatures (averaged across

252   all days of each survey period) were not different between years (2010, 19.2 C ± 0.69 SE; 2012,

253   18.8 C ± 0.88 SE; two-tailed t-test, p = 0.74). Furthermore, in the two weeks prior to our surveys,

254   maximum daily air temperatures were warmer in 2012 (18.8 C ± 1.08 SE) than in 2010 (15.2 C ±

255   0.65 SE; two-tailed t-test, p < 0.001). Air temperatures were also warmer in the winter

256   (beginning of December to end of February) preceding the post-removal survey than the winter

257   preceding the pre-removal survey (2009–2010, 13.4 C ± 0.36 SE; 2011-2012, 15.5 C ± 0.30 SE;

258   two-tailed t-test, p < 0.0001). Colder water temperatures may thus have contributed to the lower

259   overall turtle basking we observed in 2012, but this effect might have been modulated by warmer

260   air temperatures prior to our 2012 surveys.

261

262   *Effects of RES Removal on Turtle Basking*

263 The interaction between removal treatment and distance from the west end of the UCD

264 Arboretum was not significant (p = 0.18) and was removed from the model. Both treatment (p <

265 0.0001) and distance from the west end (p < 0.0001) were retained (cR^2 = 0.31, mR^2 = 0.31).

266 The non-significant interaction indicates the removal did not change the ratio of

267 WPT:RES basking distributions across the UCD Arboretum. Both pre- and post-removal, the

268 basking distribution of turtles was WPT-biased in the west end and RES-biased in the east end

269 (Fig 4). However, the proportion of basking individuals that were WPT increased from 30.5%

270 pre-removal to 41.3% post-removal (p < 0.0001; Tukey's post-hoc test, function 'glht', package

271 "multcomp"). Individual binomial GLMMs for each basking site showed removal treatment

272 effects on the WPT:RES basking ratio for site Q (p = 0.002, 9% WPT to 55% WPT; Fig 3) and a

273 marginal effect for site O (p = 0.09, 30% WPT to 75% WPT; Fig 3). All other individual basking

274 sites showed no differences (all p > 0.1).

275 Pre-removal, the WPT basking distribution declined from west to east, and post-removal

276 WPT basking had a relatively flat distribution (Fig 5). We detected a shift in the absolute basking

277 distribution of WPT with a significant interaction between removal treatment and distance from

278 the west end (Poisson GLMM, p = 0.012, cR^2 = 0.23, mR^2 = 0.06). Individual GLMMs for each

279 year indicated that distance from the west end was significantly associated with WPT basking

280 abundance in the pre-removal year (p < 0.012, cR^2 = 0.27, mR^2 = 0.03) but not in the post-

281 removal year (p = 0.55).

282 WPT predominantly used the same basking sites post-removal but showed a more even

283 distribution across basking sites, with more basking activity at two center-east sites (Q and X)

284 compared to before the RES removal (Fig 3A, B). Contingency table analyses showed that sites

285 Q (p = 0.01) and X (p = 0.001) encompassed larger proportions of total WPT basking

observations post-removal than pre-removal (Fig 3A, B). All other sites made up similar

287 proportions pre- and post-removal (all p > 0.1), though some sites had generally low basking

288 activity (Fig 3A, B), possibly limiting our power to detect shifts.

289   Our experimental removal of RES was associated with flatter observed distribution of

290 WPT. Even so, if more eastern sites that were dominated by RES pre-removal (e.g., sites O, P,

291 Q, and R) are also preferred WPT basking locations, then WPT should have increased basking at

292 these sites post-removal. We did not see this shift.

293   After removal, remaining RES were sparse throughout much of the UCD Arboretum and

294 concentrated in the east end (Fig 3B, 5). For RES, a Poisson GLMM indicated a significant

295 interaction between treatment and distance to the west end (p < 0.0001, $cR^2 = 0.30$, $mR^2 = 0.16$).

296 Individual GLMMs for each year showed a positive relationship between RES basking and the

297 distance to the west end pre-removal (p < 0.0001, $cR^2 = 0.27$, $mR^2 = 0.02$) and post-removal (p <

298 0.0001, $cR^2 = 0.14$, $mR^2 = 0.14$). The distance of each basking site from the west end explained

299 substantially more of the variation in RES basking abundance post-removal than pre-removal,

300 indicating that remaining RES concentrated in the east end more strongly after we removed most

301 of the RES population (Fig 3B, 5). Contingency table analyses indicated that sites E, O, P, and R

302 (Fig 3) comprised lower proportions of total RES basking observations after the removal and site

303 X (Fig 3) comprised a higher proportion (all p < 0.05). Site Q made up similar proportions of

304 total RES observations in both years (p = 0.14).

305   The RES remaining post-removal abandoned several basking sites that they previously

306 used heavily (particularly sites O and P; Fig 3) and shifted towards the east end of the UCD

307 Arboretum (e.g., site X). This result suggests that RES prefer habitat at this end of the waterway

308 and that, prior to our experiment, RES densities were high enough for intraspecific competition

309   to force many RES into less preferred areas of the waterway. Our previous work showed that

310   RES basking activity was highest at sites with shallow slopes, deeper water adjacent to the site, a

311   steel mesh (rather than concrete or dirt) substrate, and high human activity (Lambert et al. 2013).

312   Post-removal, RES basking activity was highest at the two sites (V and X) that maximized this

313   combination of variables based on 2010 surveys (Fig 4B from Lambert et al. 2013).

314

Discussion

316   Our experimental removal dramatically altered both RES and total turtle density in the

317   UCD Arboretum by eliminating over half of the turtles in the waterway. Given the high

318   population density of RES and given that we likely removed the majority of the RES population,

319   we consider our RES removal effort substantial enough to have exerted an effect on WPT if the

320   two species compete for food or basking sites. Our removal experiment offers new insights into

321   competition for basking habitats and food between introduced RES and native WPT, producing

322   four key results.

323   First, the prevalence of basking turtles at our survey sites post-removal was about 15% of

324   that pre-removal, and this reduction in basking observations was measured in both species. We

325   have no evidence that the removal of RES negatively affected the WPT population size, and a

326   follow-up trapping survey confirmed that the number of WPT present remained roughly

327   constant. Rather, it appears that the overall lower density of turtles in the UCD Arboretum

328   allowed many WPT to either shift their basking activity patterns, redistribute themselves to sites

329   that we were not monitoring, or both. Environmental differences, including cooler water

330   temperatures during our post-removal monitoring, may also explain the lower WPT basking

331   numbers, although our previous results from the same site suggest that the water temperature

332   during our basking surveys would support maximal WPT basking activity (Lambert et al. 2013).

333   It seems unlikely that differences in observers between years would have impacted these

334   findings.

335   Second, after removing RES, we found that WPT basking activity at our monitoring sites

336   shifted but did not increase at sites previously dominated by RES. Thus, we did not find evidence

337   of strong interspecific competition for those sites. Interspecific competition is greatest at higher

338   densities and the effects of an introduced competitor can similarly manifest or become most

339   pronounced when the introduced species is at high densities (Gurnell et al. 2004). Therefore,

340   competition is presumably greatest at high densities of RES (or turtles generally) and perhaps

341   influenced by the relative densities of both species. While earlier laboratory and mesocosm

342   experiments suggest introduced RES outcompete native turtles for basking sites and other

343   resources (Cadi and Joly 2003; Polo-Cavia et al. 2010; Pearson et al. 2015), our results suggest

344   more subtle effects found in complex, natural communities that are poorly predicted by

345   simplified mesocosm experiments (Skelly 2002, Winkler and Van Buskirk 2012).

346   Third, after removing most RES, remaining RES concentrated their basking at sites (V

347   and X) consistent with their previously identified preferred habitat characteristics (Lambert et al.

348   2013), suggesting that high RES densities prior to our experimental removal produced strong

349   intraspecific competition, forcing many RES to use less-preferred basking habitat.

350   Fourth, we found that removing RES led to an increase in WPT body condition,

351   suggesting that these turtle species compete for food. Whether this reflects interference

352   competition (direct interactions between the two species), exploitation competition (both species

353   indirectly competing for overlapping food resources), or a combination of the two is unclear.

354   Experimental work on RES and other native turtles suggests RES may behaviorally prevent

355 native turtles from obtaining sufficient food (Cadi and Joly 2004, Polo-Cavia et al. 2011, Pearson

356 et al. 2015), and our experimental removal may have reduced such interference if it does exist in

357 this population. However, we also removed a substantial portion of the overall turtle community

358 thereby reducing the overall pressure on food resources in the system. While differences in

359 trapping dates between the two years (earlier post-removal) may have influenced our results, we

360 think such effects are limited. The absence of an interaction between sex and treatment indicates

361 that male and female WPT responded similarly to RES removal and suggests that differences in

362 trapping date did not influence our results because of gravid females. Additionally, later trapping

363 pre-removal could have allowed WPT to gain more mass over the active season compared to

364 earlier trapping post-removal, making it hypothetically more challenging to detect a positive

365 effect of RES removal on WPT body condition. Because of this, our body condition results may

366 be a conservative estimate of body condition increase. Regardless of the mechanism, the ca. 40 g

367 average increase in body condition we detected is substantial given that all WPT in our analysis

368 pre-removal weighed under 1,100 g. To our knowledge, this result represents the first evidence

369 from wild populations that introduced RES compete with native turtles for food and that RES

370 removal can lead to improved body condition of native turtles.

371

372 *Should We Remove RES to Benefit Declining Native Turtles?*

373 A recent summary of research goals for effective conservation of WPT (Thomson et al. 2016)

374 identified the need for a clearer quantitative understanding of the impact of introduced RES.

375 Controlling invasive species is a substantial commitment that rarely eliminates the entire

376 population, particularly in situations with continual introductions (Kikillus et al. 2012, Gaeta et

377 al. 2015, Garcia-Diaz et al. 2017). Removing 177 RES from the UCD Arboretum was an

378   intensive effort requiring > 2,000 person-hours of field work across 40 days. A similar level of

379   effort would conservatively cost the California Department of Fish and Wildlife $26,000–

380    $31,000 in Scientific Aid hourly wages (L. Patterson, *pers. comm.*). While our study suggests

381   that removing RES does influence native turtle basking ecology and feeding, the potential

382   benefits with respect to short-term basking-site usage appear quantitatively modest. However,

383   the substantial increase in WPT body condition during the year following the RES removal

384   suggests that removing RES meaningfully increased resource availability for WPT. Whether

385   these returns justify the effort may well depend on several variables, including RES abundance /

386   density, attitudes of local human residents to introduced RES, disease risk (Héritier et al. 2017),

387   other potential axes of competition (e.g., nesting sites), and additional aspects of ecosystem

388   health.

389   Our results also provide evidence that RES introductions may affect native turtles simply

390   by inflating turtle densities in general (regardless of species identity). Therefore, removing RES

391   may not necessarily relieve native turtles from a dominant competitor but, rather, may relieve

392   ecological or behavioral pressures associated with high turtle densities and could conceivably

393   result in unexpected responses by native species. One such unexpected response here was the

394   substantial decrease in overall WPT basking observations after we removed over half of the

395   turtle community, a result that suggests a change in WPT behavior and habitat use that our

396   experimental design, with fixed monitoring sites, failed to capture. Unlike many other freshwater

397   turtles, WPT are aggressive baskers—threatening, biting, pushing, and ramming other turtles

398   from basking sites—and prefer to bask alone or in low numbers (Bury and Wolfheim 1973).

399   Reduced turtle densities post-removal may thus have allowed WPT to occupy other basking

400   habitats in lower numbers as is their preference. Additionally, higher WPT body condition post-

401 removal was likely influenced by there simply being fewer turtles overall competing for food in

402 the UCD Arboretum. Improved body condition may also be the result of WPT adopting preferred

403 basking behaviors, thereby improving digestive efficiency and mass gain. Future studies,

404 including dietary research, that include unmanipulated control sites, pre-removal surveys that

405 span multiple years and account for year-to-year variation, as well as a design that tracks the

406 behavior of native turtles pre- and post-RES removal (e.g., using GPS-enabled radio

407 transmitters) may better elucidate these unexpected outcomes on native turtles. Overall, our

408 analyses suggest WPT responded to RES removal in a manner consistent with interspecific

409 competition for food but inconsistent with strong interspecific competition for basking habitats,

410 implying that removing RES may well be an important management strategy in some situations.

411   Alternatively, the direct management of basking habitat may be a more generally

412 tractable conservation activity for WPT (Spinks et al. 2003, Thomson et al. 2016). In human-

413 modified waterways, removal of floating basking sites for flood control and aesthetics (Spinks et

414 al., 2003) could exacerbate competition for basking sites. Emerging research suggests that

415 experimentally-added floating logs are preferred by WPT compared to bank-side basking sites

416 and are more heavily used by WPT than RES, especially when they are isolated from human

417 activities (Cossman et al. unpubl.). Adding artificial basking sites that favor WPT, alone or in

418 combination with RES population reduction, is a simple, comparatively inexpensive

419 manipulation that should be explored in future field experiments.

420

421 *Study Limitations*

422 The primary limitations of our study center on interpreting our basking results. We expected to

423 observe fewer basking RES in the second year of study due to our intense removal effort but did

424 not expect a concomitant decline in WPT observations. It is possible that water temperature,

425 other environmental variation, or unforeseen consequences of our manipulation resulted in

426 reduced overall turtle basking activity, or (more likely to us) radical shifts in basking to new and

427 unmonitored locations, after the RES removal. Unfortunately, we cannot confidently identify

428 which factor(s) resulted in fewer WPT basking observations. Although we studied both basking

429 and feeding, we also recognize that our experiment did not address other potentially important

430 axes of competition that are important for the continued recruitment and persistence of WPT

431 populations. While we employed a before-after comparative design, the use of unmanipulated

432 control sites would have improved our ability to make stronger inferences in this study. We do

433 not believe that the lower number of WPT basking observations confounds our results because

434 our analyses of relative basking distribution differences between species and years can

435 accommodate sample size differences. Additionally, our analyses found that residual RES shifted

436 their basking in intuitive ways (i.e., towards sites with preferred characteristics), increasing

437 confidence in our results. While field experiments offer more biological realism than

438 experiments in captivity, that added complexity may also yield unexpected results, such as

439 changing a focal species' behaviors or habitat use.

440

441 **Conclusions**

442 We present the first *in situ* field manipulation testing for competition between non-native RES

443 and native turtles. Consistent with expectations based on laboratory and mesocosm studies, RES

444 removal increased WPT body condition and altered WPT basking activity. However, contrary to

445 expectations, this change in basking was not consistent with strong competition between RES

446 and WPT for individual basking sites in the UCD Arboretum. Our results offer evidence for

447 intraspecific competition for food and basking sites at high RES densities, underscore the value

448 of manipulative field experiments in studying biological invasions, and suggest that removing

449 introduced RES could be considered a useful, albeit logistically challenging, tool for managing

450 wild WPT in some contexts. We encourage other researchers to replicate our field-based

451 experiment, perhaps using control sites or multiple years of pre-removal observations. These

452 modifications to our protocol would improve the ability to interpret competition between RES

453 and native turtles and the magnitude of behavioral shifts that occur when removals lead to

454 changes in both relative and absolute turtle densities.

455

456 **Data Availability**: Basking and body size data are available in the electronic supplementary

457 material.

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462

463

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Figure 1

The UC Davis Arboretum waterway, turtle basking sites, and basking turtles.

Map (A) of the UC Davis Arboretum outlined with a dashed white line with the waterway in blue and turtle basking sites displayed as white-rimmed red circles. Seen basking (B) are a native western pond turtle and an introduced red-eared slider side-by-side in the Arboretum. Map data © Bing 2019. Photo credit Max Lambert.

Figure 2

Native western pond turtle (WPT) body condition before and after introduced red-eared slider (RES) removal.

Body condition is shown as the residuals of body mass regressed against plastron length. Individual WPT varied in their body condition response to introduced RES removal (A) but body condition generally improved. On average (B) WPT are 39.80 g heavier after RES removal. Boxplot hinges show the 25th and 75th body condition percentiles, whiskers show the extent of data within 1.5 times the interquartile range, and the center line is the median for each treatment year pre- and post-removal.
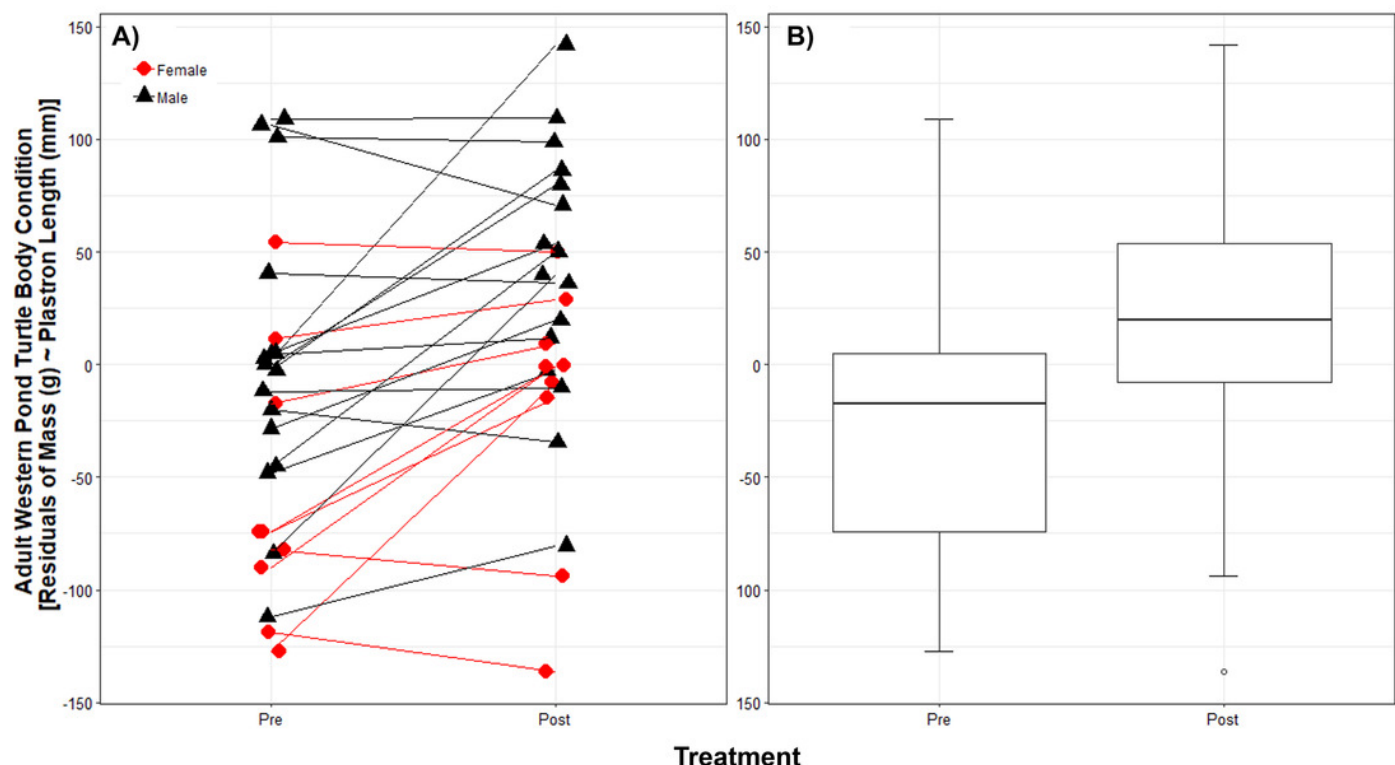
Figure 3

Cumulative basking observations of native WPT (*Emys*) and introduced RES (*Trachemys*).

Basking observations before (A) and after (B) the RES removal are arrayed along a west-east gradient in the UCD Arboretum. Letters under the x-axis are basking site identifiers. Note the y-axes are on different scales in the two panels.
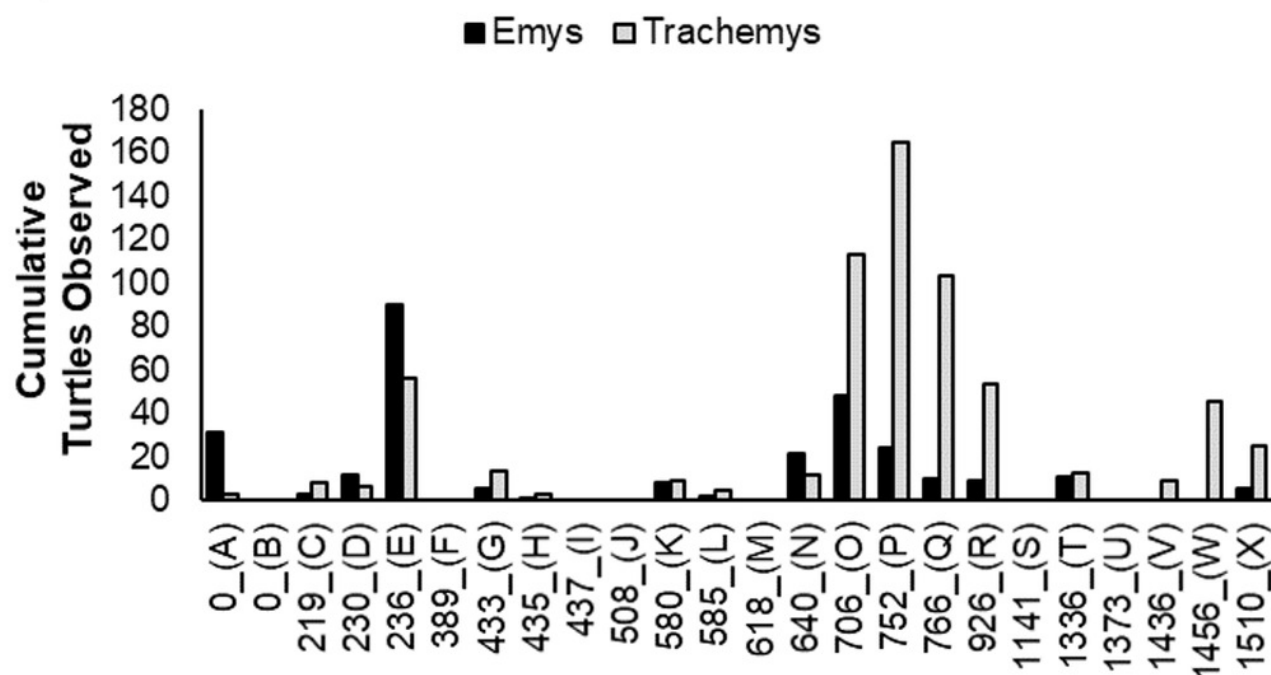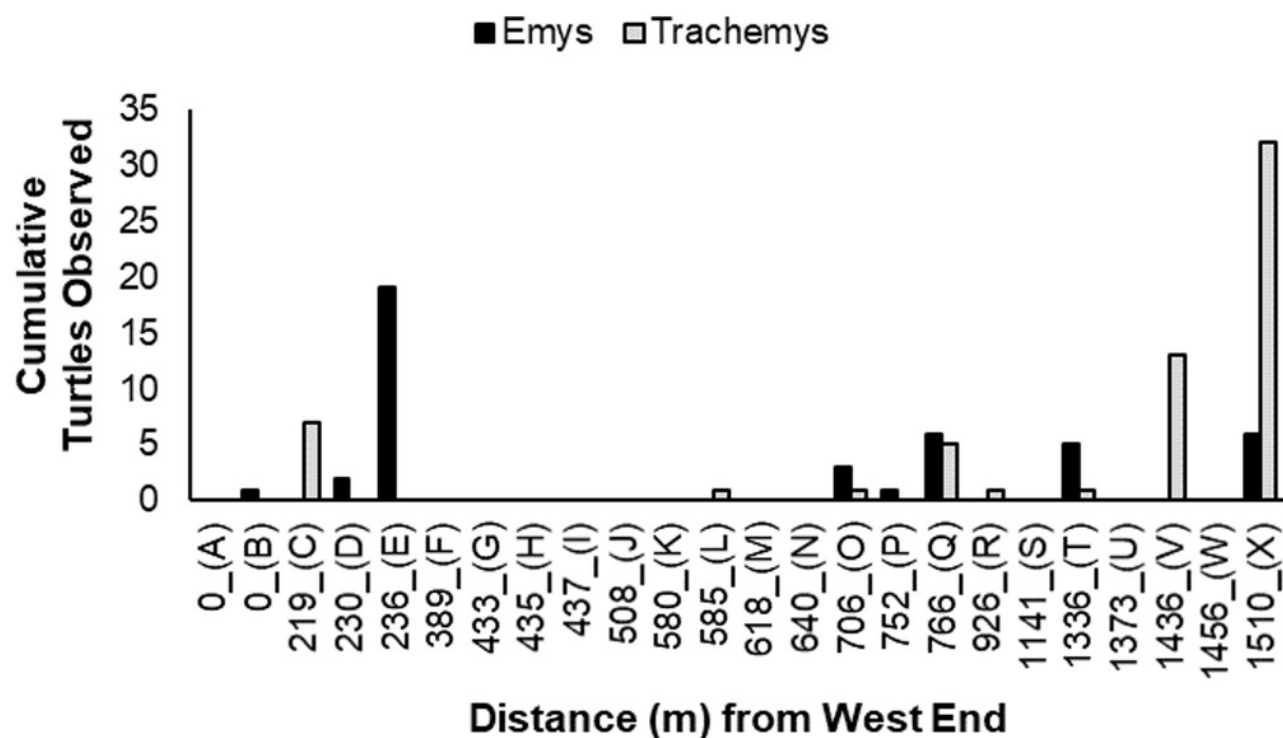
Figure 4

Relative basking distribution of native WPT (Emys) to introduced RES (Trachemys) basking across the UCD Arboretum waterway pre- and post-removal.

Curves are the modeled ratios of WPT to RES basking along a west-east gradient in the UCD Arboretum pre- and post-removal (black and red curves, respectively). Models are for the relative basking distribution of the two species and account for the binary nature of these data and variation in sample sizes across basking sites and survey dates. The ratio of WPT to RES basking along the waterway was similarly WPT-biased in the west and RES-biased in the east in both years. WPT basking observations were higher after the RES removal.
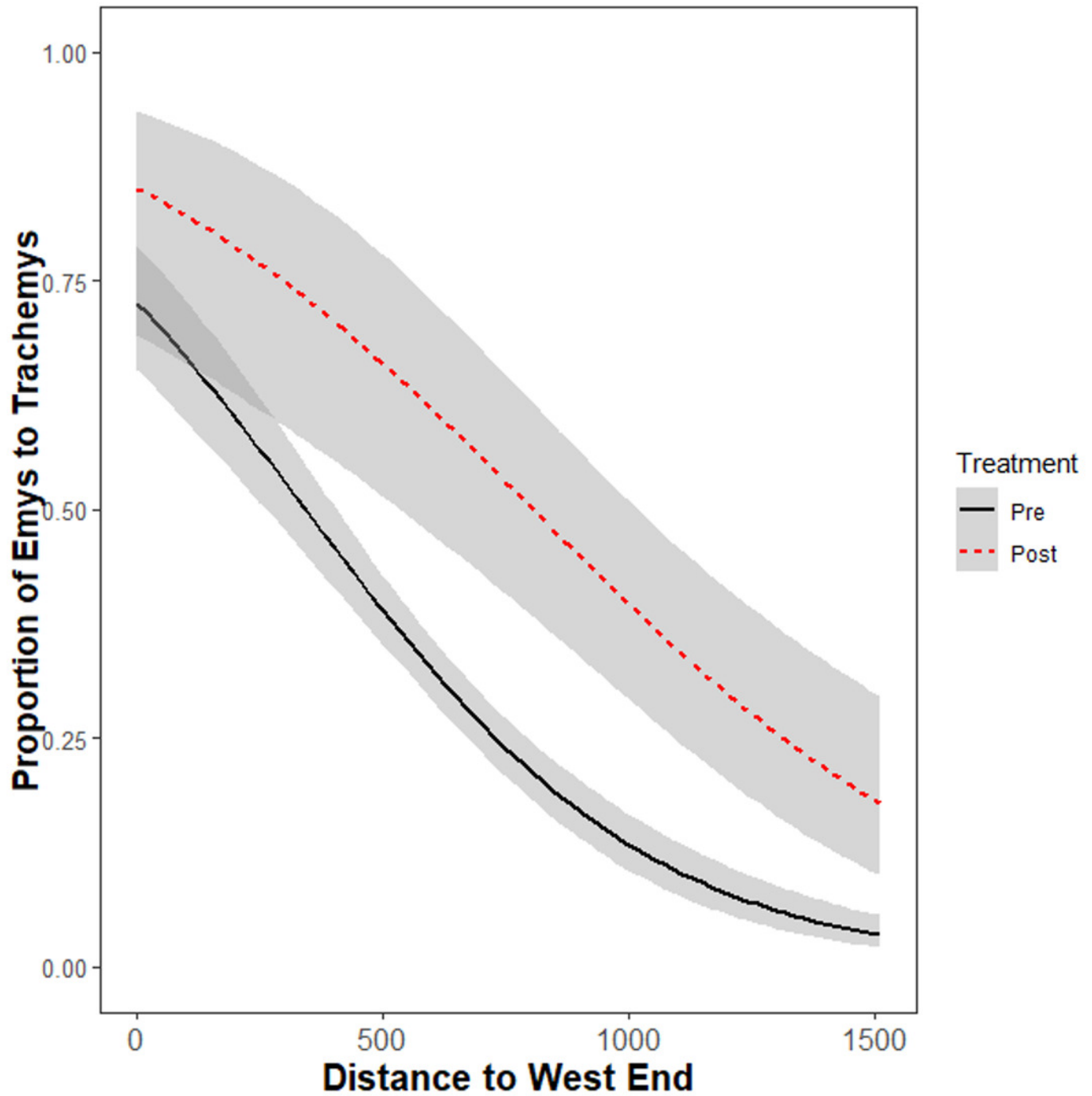
Figure 5

The total number of native WPT (*Emys*) and introduced RES (*Trachemys*) basking along the UCD Arboretum.

Curves are the modeled daily number of WPT and RES along a west-east gradient in the UCD Arboretum pre- and post-removal (black and red curves, respectively). WPT (A) displayed a more even basking distribution after the RES removal, and RES (B) concentrated basking activity towards the east end of the Arboretum after most of their population was removed.