

1 **Final word count: 5732**

2 **Fire severity drives variation in post-fire recruitment and residual seed bank size of *Acacia* species**

3

4 Harrison D. Palmer^{1,#*}, Andrew J. Denham^{1,2} and Mark K. J. Ooi³

5

6 ¹ Centre for Sustainable Ecosystem Solutions, School of Biological Sciences, University of

7 Wollongong, Wollongong, NSW 2522, Australia

8 ²Ecosystem Management Science, Office of Environment and Heritage (NSW), Hurstville, NSW 2220,

9 Australia

10 ³Centre for Ecosystem Science, School of Biological Earth and Environmental Sciences, University of

11 New South Wales, UNSW Sydney, NSW 2052, Australia

12 # Current address: Centre for Mine Site Restoration, Curtin University, Perth, WA 6102, Australia

13

14 *Corresponding author. E-mail: harrison.palmer@postgrad.curtin.edu.au

15 Orcid numbers: Harrison Palmer - orcid.org/0000-0002-0151-1158

16 Andrew Denham - orcid.org/0000-0002-8375-2747

17 Mark Ooi - orcid.org/0000-0002-3046-0417

18

19 Key words: physical dormancy; fire severity; heat shock; *Acacia*; recruitment; residual seed bank;

20 obligate seeder; land management

21 Running head: Fire severity affects *Acacia* recruitment

22 **Abstract**

23 Very high severity fires are a component of many fire-prone ecosystems, yet are often viewed as
24 detrimental to vegetation. However, species in such systems are likely to have adapted to persist
25 under a fire regime that includes high severity fires. We examined how fire severity affects post-fire
26 recruitment and residual seed banks of *Acacia* species and whether severity may affect plant
27 responses to fire intervals. Nine sites of either high or low burn severity were identified after a large-
28 scale mixed-severity fire in Warrumbungle National Park, south-eastern Australia. Transects were
29 used to sample above-ground woody plant density. Seed bank size was surveyed by soil extraction
30 from two depths and manual searching for seeds. Residual soil seed bank and recruitment were
31 compared across the two burn severities. *Acacia* seedling density was higher in areas burnt at high
32 severity, indicating that increased severity triggers increased germination from the seed bank. Size
33 of residual seed bank was smaller after high severity fire, but varied between species, with few
34 *Acacia cheelii* seeds remaining despite high above-ground abundance. In contrast, *A. penninervis*
35 retained a small residual seed bank. There was little evidence of negative effects on populations of
36 *Acacia* species after high severity burns. However, we found that high fire severity may impact on
37 the ability of a species to persist in response to a subsequent short fire-interval. Fire management
38 for maintaining biodiversity needs to consider other key aspects of the fire regime, including severity
39 and season, rather than focusing solely on fire frequency.

40 **Introduction**

41 In order to predict the response of plant populations and their ability to persist under altered fire
42 regimes, we need to understand the mechanisms that determine the magnitude and spatial and
43 temporal patterns of post-fire recovery (Whelan 1995; Bond and van Wilgen 1996). This requires
44 consideration of many interacting factors, from key life-history traits to variation in fire conditions.
45 For many species from fire-prone habitats, particularly obligate seeders (i.e. those that are killed by
46 fire and depend on recovery from seeds), the recovery of plant populations depends on the pre-fire
47 abundance and distribution of seeds in a seed bank, as well as the fire response and resilience of
48 their seed bank. Seed bank dynamics are therefore a critical determinant of population persistence.

49 In fire-prone regions, seedling emergence is related to fire-cued germination and contributes to the
50 magnitude of recruitment, while the number of seeds remaining dormant (a post-fire residual seed
51 bank) can contribute to a bet-hedging capacity, ensuring seeds are retained in the event of a lost
52 seedling cohort (Whelan 1995; Bond and van Wilgen 1996; Auld and Denham 2006). Seed dormancy
53 is one mechanism that influences both these outcomes, as it assists in maintaining a seed bank for
54 long periods of time (Auld et al. 2000; Fenner and Thompson 2005; Ooi et al. 2007). Seed mortality
55 resulting from fire can affect both the above and below ground population, resulting in lower levels
56 of recruitment and a reduced residual seed bank. High seed mortality could compromise the
57 capacity of a population to effectively recruit or retain a residual seed bank large enough to produce
58 a subsequent seedling flush if a fire occurs soon after the first (i.e. increased fire frequency) (Auld
59 and Denham 2006; Auld et al. 2007).

60 Many obligate seeding species have seeds with physical dormancy (also known as hard-seeded
61 species), a dominant dormancy type in fire-prone ecosystems (Merritt et al. 2007; Ooi 2007), which
62 is controlled by an impermeable seed coat. Physical dormancy is usually broken by high, short
63 duration temperatures experienced during the passage of fire. For successful recruitment to occur,
64 the seed bank must be heated to temperatures high enough to break the dormancy of such species,

65 but not so high as to cause excessive levels of seed mortality. The general pattern for species from
66 many fire-prone regions around the world is that germination increases in response to increasing
67 heat shock temperatures for physically dormant species (Keeley and Meyers 1985; Jeffery et al.
68 1988; Auld and O'Connell 1991; Herranz et al. 1998). In the field, this means that in areas where
69 higher temperatures are generated in the soil during fire, a larger proportion of the seed bank would
70 be exposed to dormancy-breaking thresholds (Bradstock and Auld 1995; Odion and Davis 2000; Ooi
71 et al. 2014), resulting in a potentially higher abundance of seedlings and a greater diversity of
72 species. However, once a high temperature threshold is exceeded, heat will cause seed mortality.
73 Heat transfer into soil is dependent on the characteristics of the fire (e.g. energy release,
74 temperature and duration), composition of litter (Bradstock et al. 1992; Bradstock and Auld 1995)
75 and thermal properties of the soil, which is influenced by its composition, density and moisture
76 content (Van Wijk 1963; Abu-Hamdeh and Reeder 2000; Stoof et al. 2011). Exposure of seeds to
77 dormancy-breaking or mortality thresholds is also dependent on the depth that seeds occur in the
78 soil, with soil temperatures attenuating steeply with soil depth (Bradstock et al. 1992; Bradstock and
79 Auld 1995; Penman and Towerton 2008). Across regions with similar soil characteristics and litter
80 composition, variation in fire intensity is likely to be a key determinant of recruitment response and
81 seed mortality of physically dormant species.

82 Fire severity, a measure of the physical impact of fire on vegetation and therefore a correlate of
83 intensity (Keeley 2009), is a key component of the fire regime. However, while several studies have
84 reported on the vegetation response to varying fire severity, including survival, structure and
85 magnitude of population recovery (e.g. Moreno and Oechel 1991; Morrison et al. 1992; Thaxton and
86 Platt 2006), few have related this to the mechanisms driving these responses, such as temperature-
87 related seed dormancy. Those that have are often focused on the potential negative effects of very
88 cool fires on germination and recruitment (e.g. Bradstock et al. 1992; Bradstock and Auld 1995; Ooi
89 et al. 2014). Very high severity fires are an inherent component of many fire-prone ecosystems, yet
90 are often considered detrimental to vegetation recovery and maintenance of biodiversity (e.g. Vivian

91 et al. 2008; DellaSala et al. 2015). This is despite the fact that the majority of plant species in fire-
92 prone vegetation are likely to have adapted to persist under a fire regime that includes high severity
93 fires. Quantification of the effects of very high fire severity on the persistence of plant populations is
94 therefore required.

95 One basis for the perceived negative impact of extremely severe fires in naturally fire-prone
96 systems, is the assumption that soil temperatures cause excessive seed mortality, to a much greater
97 extent than during more moderate fire events (Bradstock and Auld 1995), potentially reducing both
98 the magnitude of emergence and the size of the residual seed bank. Under this assumption,
99 recruitment after extremely severe fire would depend on the resilience of seeds to high
100 temperatures and/or the depth of burial of seeds. Soil offers highly effective insulation, and
101 temperatures decrease considerably with increasing depth (Bradstock et al. 1992; Tozer 1998;
102 Penman and Towerton 2008). During fire events, seeds that are closer to the surface, where soil
103 heating is greatest, are much more likely to be exposed to dormancy-breaking temperatures, but
104 may be at greater risk of mortality. The size of the residual soil seed bank, which can bet-hedge
105 against a loss of a seedling cohort, would also be affected by increased seed mortality, potentially
106 retaining fewer seeds and at greater depths (Auld and Denham 2006). Under extreme fire severity, it
107 is therefore likely that a greater proportion of seeds will die, but seeds buried deeper will have less
108 chance of suffering mortality. Thus, factors such as seed size, which can determine the ability to
109 germinate successfully from depth (Bond et al. 1999, Auld and Denham 2005), and depth of burial
110 will interact with fire severity to determine recruitment potential.

111 In this study, we investigated how fire severity impacts both recruitment and the residual soil seed
112 bank of the dominant understorey species in a *Eucalyptus-Callitris* forest in south-eastern Australia.
113 For the two key canopy species groups, high intensity, severe fire kills *Callitris* species which must
114 then recruit from a canopy seed bank or from seeds dispersing into the burnt area, and causes above
115 ground mortality in *Eucalyptus* requiring resprouting from protected meristems below ground.

116 However, these species are resilient to lower severity fires (Cohn et al. 2011, Denham et al. 2016).
117 Extremely high severity fires therefore retard the rate of recovery of these canopy species. The
118 response to varying fire intensity of many of the understorey species is less well understood.
119 Research identifying how fire regime shifts in temperate fire-prone regions may drive vegetation
120 change has recently been highlighted as an important knowledge gap (e.g. see Fairman et al. 2016).
121 A key aim of our study is to therefore assess how extreme fire events affect recruitment and
122 persistence of understorey species in this system, where the majority of plant diversity occurs.

123 Our focal species are a dominant component of the understorey, all occurring within the genus
124 *Acacia*, with heat responding physically dormant seeds. We considered two hypotheses; firstly, that
125 higher fire severity will increase recruitment in *Acacia* species, and secondly, that higher fire severity
126 would reduce the abundance of seeds in the residual soil seed bank, potentially limiting the bet-
127 hedging ability of these populations. To test these hypotheses, we quantified plant recruitment
128 above-ground and the density of the corresponding residual soil seed bank below-ground in areas
129 burnt by either extremely high or relatively low severity fire. We incorporated depth as a factor in
130 our experiments to further assess the impacts of fire severity on residual seed bank densities. More
131 specifically, we addressed the following questions:

- 132 1. Does fire severity impact recruitment of *Acacia* species from the seed bank and
133 does the effect differ among species?
- 134 2. Does fire severity affect the residual seed bank, and if so, is this related to seed
135 burial depth?
- 136 3. Is there evidence for negative effects of extremely high fire severity on the
137 persistence of populations of fire-prone physically dormant species?

138

139 **Methods**

140 *Study Area*

141 The Warrumbungle National Park (WNP) covers approximately 23 000ha in the upper reaches of the
142 Castlereagh River catchment, south eastern Australia (31.29°S, 149.01°E). The region is
143 topographically complex (elevation range 400 - 1200 m), with soils derived from basalts, trachyte
144 and pyroclastic flows and from pre-volcanic sandstones. Summers are warm to hot (mean maximum
145 temperature 23.7°C), while winters are mild (mean minimum temperature 7.4°C), with an average
146 annual rainfall of 750 mm (mostly in summer, though soil remains moist in winter).

147 The vegetation consists mostly of open *Eucalyptus-Callitris* forest (North-west Slopes Dry Sclerophyll
148 Woodlands and Western Slopes Dry Sclerophyll Forests) (Keith 2004), with an understorey of
149 sclerophyllous shrubs in low soil fertility areas, while grasses and forbs dominate high soil fertility
150 areas (Denham et al. 2016). Canopy dominants vary among habitats, but *Callitris glaucophylla*,
151 *Eucalyptus albens* and *E. crebra* are the predominant tree species present in most communities
152 (Hunter 2008), and are 20-30 m tall. Obligate seeder species with physically dormant seeds
153 dominate the understorey of most vegetation communities in the region, with genera such as *Acacia*
154 and *Dodonaea* being very common. While these understorey species re-establish rapidly after fire,
155 *Callitris* populations may take over ten years to recover from a severe fire event, as population
156 regeneration depends on relatively slow growing seedlings (Denham et al. 2016).

157 The recorded fire history of the WNP shows that there have been several relatively small fires
158 scattered over the past 70 years, but that a majority of the park has not been burned for 40 years
159 (OEH unpublished data, Storey et al. 2016). The Wambelong Fire, which occurred in 2013, is the
160 largest and most severe fire in the recorded history of the park and is the focus of this study. During
161 extreme fire conditions in January 2013, the fire burnt over 39 000 ha of vegetation within 24 hours.
162 Following the initial conflagration, the fire continued to burn in low-moderate fire conditions for
163 several weeks, burning another 17 000 ha before being extinguished on the 21 Feb. Overall,
164 approximately 84% of WNP was burnt, with 13% burnt at low severity, indicated by scorched low

165 shrubs but tree canopies remaining unburnt. Around 64% of the park was burnt at high or extremely
166 high intensity, with severity class in these areas based on top-killed trees and consumption of all
167 vegetation and canopy (Denham et al. 2016).

168

169 *Site selection*

170 Potential study areas were initially selected based on remote sensing classification, and severity
171 subsequently confirmed by field assessment. Sites were only considered if individuals of the study
172 species were present. Sites were categorised as being burnt at either low or high severity, which was
173 a surrogate for fire intensity. Fire intensity was classed according to Chafer et al. (2004), who
174 considered the amount of energy expended and type of burn. Classes were low-moderate (fire
175 intensity $<500 \text{ kWm}^{-1}$, surface and low shrub fire) and high-extreme ($500\text{-}70\,000 \text{ kWm}^{-1}$, tall shrub
176 fire with tree canopy scorch to total canopy consumption). Field assessment of severity was based
177 on height of canopy scorch/consumption and the proportion of litter consumption. High severity
178 sites consisted of areas where all litter, shrubs and most tree canopies (mainly *E. crebra*, *E. albens*
179 and *C. glaucophylla* canopies $>15 \text{ m}$) were consumed by fire, and only tree skeletons remained. At
180 low severity sites, shrubs and litter were consumed, but tree canopies remained intact or partially
181 burnt and *Eucalyptus* trees were observed resprouting from epicormic buds. Upon identification of
182 suitable sites of high or low severity, a $20 \times 20 \text{ m}$ plot was established, within which surveys were
183 conducted (See Table 1 for a description of sites).

184 Five sites burnt at low severity and four sites burnt at high severity were identified across different
185 areas of the park, accounting for spatial variation of population distribution and fire characteristics.
186 *Acacia* species within these sites were identified and targeted for the study (Table 2). Topography
187 was similar across all sites, being either flat or on gentle slopes ($<10\%$ slope). The post-fire density of
188 vegetation in the shrub layer varied among sites, ranging from scattered individuals to extremely
189 dense (Table 1).

190

191 *Recruitment response to fire severity*

Commented [HDP1]: Rewrote this whole section

192 Field work to assess recruitment via density measurements was conducted in August 2015,
193 approximately two and a half years after the fire. Density of post-fire recruits and surviving adults (if
194 any) of woody species was calculated using transect surveys. Three 10m belt transects were laid out
195 from along the east and west edges of each plot, with the third following the same orientation but
196 through the centre of plots. All woody stemmed plants within 1m either side of each transect were
197 identified and counted, giving a total density for a surveyed area of 60 m². Density measurements
198 (plants per m²) were calculated separately for each transect for total woody vegetation density and
199 3 subgroups: density of *Acacia* species and the two dominant *Acacia* species. Density calculations of
200 *A. cheelii* excluded sites where *A. penninervis* was dominant (and *A. cheelii* absent) and vice versa,
201 including sites where they both were present.

202

203 *Residual seed bank size*

204 The residual seed bank was surveyed while the recovering vegetation was at the juvenile stage, and
205 thus had not contributed to the seed bank prior to sampling. This allowed us to survey only the
206 residual seed bank that persisted through the 2013 fire. After vegetation surveys were completed,
207 stratified random sampling was undertaken to select eight 30 x 30 cm seed bank samples at each
208 site. Seed bank sample plots were stratified by the number of above ground *Acacia* seedlings
209 present, then randomly selecting their location in the site. The eight samples comprised two each
210 with 0, 1-2, 3-4 or 5+ seedlings. Once samples were located, seedlings were counted and removed.
211 Soil from each sample was then removed to a depth of 5 cm, bagged and labelled and then to a
212 depth of 10 cm (total volume of 900 cm³ per sample), ensuring soil from depth classes was kept
213 separate. Soil was then taken to the laboratory, allowed to dry, and sieved to separate coarse and

214 fine particles to ease the sorting process. The samples were sorted by hand to find seeds of all
215 *Acacia* species, which were removed from the soil, identified and recorded. The mean number of
216 seeds at each depth class was calculated for each severity class, using samples as replicates.

217

218 *Data analysis*

219 To compare the density of above ground species among sites burnt at different severities, we used a
220 Generalised Linear Mixed Model (GLMM) with Poisson distribution. Fire severity was included as a

221 fixed factor, while the random factor included transects nested within sites. Models were fitted to

222 compare the plant density of woody stemmed species and the density of *Acacia* (the dominant

223 component of all sites) between high and low severity burn areas. Two further analyses explored the

224 density of the two dominant species, *A. cheelii* and *A. penninervis*, using only the sites in which each

225 target species occurred. *Acacia cheelii* was dominant in occurred in 4 sites, and *Acacia penninervis*

226 dominant in 3, with both species occurring in only 2 of the 9 sites (see Table 1.)

227 To assess the effect of fire severity on residual seed bank density and whether this varied with

228 depth, a two-factor Generalised Linear Model (GLM) with Poisson distribution was used, with the

229 number of seeds recorded of all *Acacia* species as the dependent variable, and fire severity (high

230 versus low) and depth class (0-5 cm and 5-10 cm) as the two factors. To examine the difference in

231 residual seed bank density between *A. cheelii* and *A. penninervis*, depth classes were pooled and a

232 two-factor GLM was used. To test whether seeds were abundant in areas that we didn't sample, we

233 used logistic regression to model the likelihood of seeds being present in the residual seed bank in

234 relation to the number of seedlings present, fire severity, and species (*A. cheelii* or *A. penninervis*).

235

236 **Results**

237 *Recruitment response to fire severity*

Deleted:

Commented [HDP2]: I think this change makes this clearer, and as we didn't present results on 'number of pd species' I simplified it so it just states that we measured the density of woody plant species, acacias grouped, and the individual species

Deleted: number of individuals of all plant species at

Deleted: , and the number of physically dormant species (hard-seeded and known to have a heat-induced response) only at each site.

243 The mean density of live stems (all species combined) in high severity burn areas was 8.25
244 individuals per m² compared to only 2.6 individuals per m² in the low severity burn areas (Fig. 1,
245 Table 1). All four comparisons with GLMMs of above-ground vegetation demonstrated that high
246 severity burn sites had a higher density of recruitment than low severity sites (although *A.*
247 *penninervis* was only near significant; Table 3). This consistent relationship indicated that *Acacia*
248 species, and in particular *A. cheelii* and *A. penninervis*, drove the overall patterns of understorey
249 shrub response. *Acacia cheelii* and *A. penninervis* were the dominant components in the studied
250 sites, evident by their high density in both low and high severity burn areas when compared to other
251 surveyed plant species (Fig. 2).

Formatted: Highlight

Deleted: abundance

252

253 Residual seed bank size

254 The number of seeds recovered from soil seed bank samples was very low across all sites (Fig. 3).
255 Nevertheless, there was a significant effect of fire severity ($df = 1$, $\chi^2 = 4.993$, $P = 0.025$) on seed bank
256 density, with lower densities of seeds in areas burnt at high severity (0-7 seeds per m²) than at low
257 severity (0-13 seeds per m²). While there was no significant effect of depth ($df = 1$, $\chi^2 = 1.625$, $P =$
258 0.202), there was a trend for more seeds at greater depths at both fire severity classes.

259 Comparison of the residual seed banks of the two dominant *Acacia* species in relation to fire severity
260 uncovered striking differences. *Acacia penninervis* seed density was much higher than for *A. cheelii*
261 (Fig. 4). There was a significant interaction between species and severity ($df = 1$, $\chi^2 = 8.033$, $P =$
262 0.005), indicating that high severity fire significantly reduced the size of the residual seed bank, but
263 only for *A. penninervis*. *Acacia cheelii* seed density was negligible regardless of fire severity (Fig. 4).

264 Neither fire severity nor number of seedlings present significantly influenced the likelihood of finding
265 a residual seed bank according to the logistic regression model. However, species was significant,
266 such that the likelihood of finding seeds in the soil was approximately five times greater for *A.*

268 *penninervis* than for *A. cheelii* (*A. cheelii* odds ratio 0.21, 95% CI 0.06-0.81). There was a greater
269 proportion of samples that had seeds in samples with more seedlings of *A. penninervis*, but this
270 trend was not significant (Fig. 5). The low proportion of samples with seeds where there were no
271 seedlings (less than 20%) suggests that there are unlikely to be large numbers of seeds in areas
272 where we did not sample.

273

274 **Discussion**

275 High severity fire resulted in a greater recruitment response of the dominant physically dormant
276 *Acacia* species than low severity fire, with *Acacia* species dominating the mid-storey vegetation
277 response to fire. The corollary of this response is that high severity fire resulted in a greatly reduced
278 residual seed bank. A combination of large seeds and a relatively large store of seeds in the soil, in
279 conjunction with adequate soil heating from the fire, is likely to contribute to promoting the rapid
280 establishment and growth of these species. The known heat response for breaking dormancy of
281 *Acacia* seeds (Auld and O'Connell 1991; Ooi et al. 2014) provides a clear mechanism to explain the
282 patterns of recruitment observed, with the dense recovery in high severity areas indicating that
283 more seeds were exposed to temperatures high enough to break dormancy. These findings also
284 suggest that high severity fire does not necessarily result in negative outcomes for species with
285 physically dormant seeds, and that species are adapted to such events. Results from our assessment
286 of residual seed banks demonstrate that some risk-spreading capacity can be maintained after high
287 severity fires. However, the interaction of fire severity and fire frequency is important when trying to
288 determine appropriate fire intervals for the persistence of a species.

289 The above-ground recruitment recorded in our study is consistent with the hypothesis that higher
290 severity fire will produce hotter soil temperatures and subsequently greater recruitment of
291 physically dormant heat-responsive species (Ooi et al. 2014; Wright et al. 2016). Across regions with
292 similar litter composition, it is clear that other factors, such as air temperature and soil moisture,

293 contribute to fire intensity and therefore soil temperature (Whelan 1995; Stoof et al. 2011; Mondal
294 and Sakumar 2014). Higher levels of recruitment 2.5 years post-fire in areas of high severity,
295 compared to that in low severity areas, indicate that this is a key driver of the magnitude of
296 recruitment of physically dormant species, as suggested by some other studies (e.g. Knox and
297 Morrison 2005), including at other sites in relation to the same fire (Gordon et al. 2017). However,
298 few studies have quantified this, or incorporated the dynamics and role of residual seed banks in
299 determining persistence.

300 Extreme fire events, such as the one studied here, increase soil temperatures to very high levels, and
301 therefore also increase the likelihood of greater amounts of seed mortality. However, despite this,
302 high levels of above-ground recruitment were observed in higher severity burnt areas. This suggests
303 that seed mortality alone, as a result of hotter fires, is unlikely to limit recruitment in these fire-
304 adapted species. While lethal temperatures could cause greater seed mortality, this is likely to be
305 restricted to the upper few centimetres due to the insulating effects of soil. Suitable dormancy-
306 breaking temperatures would therefore shift further down the soil profile, and germination (rather
307 than mortality) would be possible for depths beyond the first few centimetres. Seed size would
308 subsequently be a critical determinant of recruitment success under increasingly severe fire,
309 because smaller seeds are unable to emerge from greater depths (Bond et al. 1999). *Acacia* species
310 have large seeds in comparison to most other species within the study region, and they would have
311 some capacity to emerge from depths of at least 5 cm (Liyanage and Ooi in review). However, the
312 fact that there is a range of seed sizes, even within our study group (9.2 – 72.8 mg, Table 2), means
313 that there could still be relative differences in response. Understanding recruitment of different
314 species to fire severity therefore requires knowledge of seed dormancy-breaking temperature and
315 mortality thresholds (Ooi et al. 2014; Liyanage and Ooi 2015), along with their ability to emerge from
316 depth (Bond et al. 1999; Hanley et al. 2003). Further work is required to estimate dormancy-
317 breaking and mortality temperature thresholds for physically dormant species in fire-prone regions.

318 Fire severity had a clear effect on the size of the residual soil seed bank. For *A. penninervis*, there
319 was more than three times the number of seeds remaining in the seed bank in the low severity
320 compared to high severity areas. In contrast, there was almost no evidence of a residual seed bank
321 for *A. cheelii* or any other *Acacia* species at sites of either high or low severity, despite the high
322 abundance of above-ground recruits. Above-ground recruitment of *A. cheelii* was comparable to *A.*
323 *penninervis*, suggesting that there were either fewer *A. cheelii* seeds prior to the fire or that more
324 were killed during the fire or died prior to emergence. *Acacia cheelii* seeds have a mass less than
325 25% that of *A. penninervis*, and would have a lower probability of successful emergence if dormancy
326 was broken at depth. Individuals of larger seeded species, such as *Acacia penninervis*, would be able
327 to emerge from greater depths where they are protected from the most extreme temperatures and
328 have the resources to reach the surface.

329 We found no evidence that the size of the residual seed bank size is moderated by burial depth. In
330 contrast, Auld and Denham (2006) showed that post-fire *Acacia* soil seed banks contained fewer
331 seeds in the top 5 cm of soil than layers from 5-10 cm and 10-15 cm, indicating that heat shock cues
332 declined with depth and caused more germination and mortality close to the surface. The lack of
333 significance of burial depth in our study could be attributed to the extremely severe fire causing very
334 high soil temperatures, even in low severity burn areas, a clear demonstration of the extreme nature
335 of the event. These high soil temperatures were likely sufficient to produce seed mortality or
336 promote germination down to a depth of at least 10 cm. The residual seed bank density (0-13 seeds
337 per m²) was low in comparison to other post-fire studies. Auld et al. (2007) found approximately 21
338 seeds per m² of *Persoonia lanceolata* (an obligate seeder) in heath and woodlands in coastal New
339 South Wales, nine years after fire. Another study found that several shrub species (including *A.*
340 *suaveolens*) in open woodlands in south eastern Australia maintained sizeable seed banks through
341 low-moderate severity fires (Auld and Denham 2006). However, there has been little study regarding
342 the size of residual soil seed banks following high severity wild fires. We suggest that further studies

343 are required after natural fires to gain a clearer understanding of the role that residual seed banks
344 play.

345 Despite significant levels of above-ground recruitment of physically dormant species post-fire, the
346 limited residual seed bank we observed suggests that a short interval before a subsequent fire event
347 would threaten populations of many of the species in the study region. Frequent fire poses a risk of
348 population decline and extinction for obligate seeding species, as individuals can be killed before
349 they mature, and seed banks are not replenished (Keith 1996; Bowman et al. 2014a; Enright et al.
350 2015). Depending on the length of the juvenile period (from emergence to flowering), this can take
351 many years (Keith 1996). While *A. penninervis* has some capacity to respond to a subsequent fire in
352 the short term, due to a small residual seed bank, many other *Acacia* species are unlikely to have
353 such a buffer. Here importantly, we have demonstrated that severity of the previous fire has
354 important consequences for a species response to subsequent fire interval, similar to studies
355 investigating obligate seeders in other regions (e.g. Bowman et al. 2014b in alpine habitat). This
356 highlights that an understanding of key elements of the fire regime beyond only fire frequency,
357 including severity and season (Ooi 2010; Mackenzie et al. 2016), need to be incorporated into
358 implemented fire strategies when managing for biodiversity or the persistence of threatened
359 species.

360

361 **Acknowledgements**

362 We thank Martin Henery and Justin Collette for field assistance, Lisa Metcalfe for laboratory
363 assistance, Craig Wall and the National Parks and Wildlife Service Coonabarabran office for
364 permission to work in Warrumbungle NP and Jessica Meade for assistance with drafting figures. The
365 project was supported by funding from the NSW Office of Environment and Heritage, and as part of
366 the Australian Government's National Environmental Science Programme (NESP), Threatened
367 Species Recovery Hub (1.3).

368

369 **References**

- 370 Abu-Hamdeh NH, Reeder RC (2000) Soil thermal conductivity effects of density, moisture, salt
371 concentration, and organic matter. *Soil Sci Soc Am J*: 64:1285
- 372 Auld TD, Denham AJ (2005) A technique to estimate the pre-fire depth of burial of *Grevillea* seeds
373 using seedlings after fire. *Aust J Bot* 53: 401-405
- 374 Auld TD, Denham AJ (2006) How much seed remains in the soil after a fire? *Plant Ecol* 187: 15-24
- 375 Auld TD, Denham AJ, Turner K (2007) Dispersal and recruitment dynamics in the fleshy-fruited
376 *Persoonia lanceolata* (Proteaceae). *J Veg Sci* 18: 903-910
- 377 Auld TD, Keith DA, Bradstock RA (2000) Patterns in longevity of soil seedbanks in fire-prone
378 communities of south-eastern Australia. *Aust J Bot* 48: 539-548
- 379 Auld TD, O'Connell MA (1991) Predicting patterns of post-fire germination in 35 eastern Australian
380 Fabaceae. *Aust J Ecol* 16: 53-70
- 381 Bond WJ, Honig M, Maze KE (1999) Seed size and seedling emergence: an allometric relationship and
382 some ecological implications. *Oecologia* 120: 132-136
- 383 Bond WJ, van Wilgen BW (1996) *Fire and plants*. Chapman and Hall, London
- 384 Bowman DMJS, MacDermott HJ, Nichols SC, Murphy BP (2014a) A grass-fire cycle eliminates an
385 obligate-seeding tree in a tropical savanna. *Ecol Evol* 4: 4185-4194
- 386 Bowman DMJS, Murphy BP, Neyland DLJ, Williamson GJ, Prior LD (2014b) Abrupt fire regime change
387 may cause landscape-wide loss of mature obligate seeder forests. *Glob Change Biol* 20: 1008-
388 1015
- 389 Bradstock RA, Auld TD (1995) Soil temperatures during experimental bushfires in relation to fire
390 intensity: Consequences for legume germination and fire management in south-eastern
391 Australia. *J Appl Ecol* 32: 76-84
- 392 Bradstock RA, Auld TD, Ellis MV, Cohn JS (1992) Soil temperatures during bushfires in semi-arid,
393 mallee shrublands. *Aust J Ecol* 17: 433-440

394 Chafer CJ, Noonan M, Macnaught E (2004) The post-fire measurement of fire severity and intensity
395 in the Christmas 2001 Sydney wildfires. *Int J Wildland Fire* 13: 227-240

396 Cohn JS, Lunt ID, Ross KA, Bradstock RA (2011) How do slow-growing, fire-sensitive conifers survive
397 in flammable eucalypt woodlands? *J Veg Sci* 22: 425-435

398 DellaSala DA, Lindenmayer DB, Hanson CT, Furnish J (2015) In the aftermath of fire: Logging and
399 related actions degrade mixed- and high-severity burn areas. In: DellaSala DA, Hanson CT (eds)
400 The ecological importance of mixed-severity fires. Elsevier, pp 313-347

401 DECC (2002) NSW Flora fire response database Version 1.3. NSW Department of Environment and
402 Climate Change, Hurstville.

403 Denham AJ, Vincent BE, Clarke PJ, Auld TD (2016) Responses of tree species to a severe fire indicate
404 major structural change to *Eucalyptus-Callitris* forests. *Plant Ecol* 217: 617-629

405 Enright NJ, Fontaine JB, Bowman DMJS, Bradstock RA, Williams RJ (2015) Interval squeeze: altered
406 fire regimes and demographic responses interact to threaten woody species persistence as
407 climate changes. *Front Ecol Environ* 13: 265-272

408 Fairman TA, Nitschke CR, Bennett LT (2016) Too much, too soon? A review of the effects of
409 increasing wildfire frequency on tree mortality and regeneration in temperate eucalypt forests.
410 *Int J Wildland Fire* 25: 831-848

411 Fenner M, Thompson K (2005) The ecology of seeds. CAB International, Wallingford

412 Gibson MR, Richardson DM, Marchante E et al. (2011) Reproductive biology of Australian acacias:
413 important mediator of invasiveness? *Divers Distrib* 17: 911-933.

414 Gordon CE, Price OF, Tasker EM, Denham AJ (2017) *Acacia* shrubs respond positively to high severity
415 wildfire: Implications for conservation and fuel hazard management. *Sci Total Environ* 575: 858-
416 868

417 Hanley ME, Unna JE, Darvill B (2003) Seed size and germination response: a relationship for fire-
418 following plant species exposed to thermal shock. *Oecologia* 134: 18-22

419 Herranz JM, Ferrandis P, Martinez-Sanchez JJ (1998) Influence of heat on seed germination of seven

420 Mediterranean Leguminosae species. *Plant Ecol* 136: 95-103

421 Hunter JT (2008) Vegetation and floristics of Warrumbungle National Park. Report to NSW National
422 Parks and Wildlife Service, Coonabarabran, Australia

423 Jeffery DJ, Holmes PM, Rebelo AG (1988) Effects of dry heat on seed germination in selected
424 indigenous and alien legume species in South Africa. *S Afr J Bot* 54: 28-34

425 Keeley JE (2009) Fire intensity, fire severity and burn severity: a brief review and suggested usage.
426 *Int J Wildland Fire* 18: 116-126

427 Keeley JE, Meyers A (1985) Effect of heat on seed germination of southwestern *Yucca* species.
428 *Southwestern Nat* 30: 304-303

429 Keith DA (1996) Fire-driven extinction of plant populations: A synthesis of theory and review of
430 evidence from Australian vegetation. *P Linn Soc NSW* 116: 37-78

431 Keith DA (2004) Ocean shores to desert dunes: The native vegetation of New South Wales.
432 Department of Environment and Conservation (NSW), Hurstville, Australia

433 Knox KJE, Morrison DA (2005) Effects of inter-fire intervals on the reproductive output of resprouters
434 and obligate seeders in the Proteaceae. *Austral Ecol* 30: 407-413

435 Liyanage GS, Ooi MKJ (2015) Intra-population level variation in thresholds for physical dormancy-
436 breaking temperature. *Ann Bot-London* 116: 123-131

437 Liyanage GS, Ooi MKJ (in review) Seed size related dormancy-breaking temperature thresholds: A
438 case for the selective pressure of fire on physically dormant species. *Biol J Linn Soc*

439 Mackenzie BDE, Auld TD, Keith DA, Hui FKC, Ooi MKJ (2016) The effect of seasonal ambient
440 temperatures on fire-stimulated germination of species with physiological dormancy: A case
441 study using *Boronia* (Rutaceae). *PLOS ONE* 11: e0156142

442 Merritt DJ, Turner SR, Clarke S, Dixon KW (2007) Seed dormancy and germination stimulation
443 syndromes for Australian temperate species. *Aust J Bot* 55: 336-344

444 Mondal NS, Sukumar R (2014) Fire and soil temperatures during controlled burns in seasonally dry
445 tropical forests of southern India. *Curr Sci India* 107: 1590-1594

446 Moreno JM, Oechel WC (1991) Fire intensity effects on germination of shrubs and herbs in southern
447 California chaparral. *Ecology* 72: 1993-2004

448 Morrison DA, Auld TD, Rish S, Porter C, McClay K (1992) Patterns of testa-imposed seed dormancy in
449 native Australian legumes. *Ann Bot-London* 70: 157-163

450 Odion DC, Davis FW (2000) Fire, soil heating, and the formation of vegetation patterns in chaparral.
451 *Ecol Monogr* 70: 149-169

452 Ooi MKJ (2007) Dormancy classification and potential dormancy-breaking cues for shrub species
453 from fire-prone south-eastern Australia. In: Adkins SA, Ashmore SE, Navie SC (eds) *Seeds:
454 Biology, development and ecology*. CAB International, pp 205-216

455 Ooi MKJ (2010) Delayed emergence and post-fire recruitment success: effects of seasonal
456 germination, fire season and dormancy type. *Aust J Bot* 58: 248-256

457 Ooi MKJ, Auld TD, Whelan RJ (2007) Distinguishing between persistence and dormancy in soil seed
458 banks of three shrub species from fire-prone southeastern Australia. *J Veg Sci* 18: 405-412

459 Ooi MKJ, Denham AJ, Santana VM, Auld TD (2014) Temperature thresholds of physically dormant
460 seeds and plant functional response to fire: variation among species and relative impact of
461 climate change. *Ecol Evol* 4: 656-671

462 Penman TD, Towerton AL (2008) Soil temperatures during autumn prescribed burning: Implications
463 for the germination of fire responsive species? *Int J Wildland Fire* 17: 572-578

464 Stoof CR, De Kort A, Bishop TFA, Moore D, Wesseling JG, Ritsema CJ (2011) How rock fragments and
465 moisture affect soil temperatures during fire. *Soil Sci Soc Am J* 75: 1133-1143

466 Storey M, Price O, Tasker E (2016) The role of weather, past fire and topography in crown fire
467 occurrence in eastern Australia. *Int J Wildland Fire* 25: 1048-1060

468 Thaxton JM, Platt WJ (2006) Small-scale fuel variation alters fire intensity and shrub abundance in a
469 pine savanna. *Ecology* 87: 1331-1337

470 Tozer MG (1998) Distribution of the soil seedbank and influence of fire on seedling emergence in
471 *Acacia saligna* growing on the central coast of New South Wales. *Aust J Bot* 46: 743-755

472 Van Wijk WR (1963) Physics of plant environment. North-Holland Publishing Company, Amsterdam
473 Vivian LM, Cary GJ, Bradstock RA, Gill AM (2008) Influence of fire severity on the regeneration,
474 recruitment and distribution of eucalypts in the Cotter River Catchment, Australian Capital
475 Territory. Austral Ecol 33: 55-67
476 Whelan RJ (1995) The ecology of fire. Cambridge University Press, Cambridge
477 Wright BR, Latz PK, Zuur AF (2016) Fire severity mediates seedling recruitment patterns in slender
478 mulga (*Acacia aptaneura*), a fire-sensitive Australian desert shrub with heat-stimulated
479 germination. Plant Ecol 217: 789-800
480
481

482 **Table 1.** Fire severity, dominant *Acacia* species (*A. cheelii*, *A. penninervis* or 'both' where present in
483 similar numbers) and mean post-fire density of stems of all woody species for study sites.

484

Site	Severity	Dominant <i>Acacia</i>	Stem density (per m ²)
1	Low	<i>A. cheelii</i>	1.62
2	Low	<i>A. cheelii</i>	1.42
3	Low	<i>A. penninervis</i>	4.83
4	Low	<i>A. penninervis</i>	2.30
5	Low	Both	2.85
6	High	<i>A. cheelii</i>	10.73
7	High	<i>A. cheelii</i>	6.42
8	High	Both	6.07
9	High	<i>A. penninervis</i>	9.77

485

486

487 **Table 2.** *Acacia* species identified in plots within the study area. Fire response categories are
 488 primarily from the NSW Flora Fire Response Database Version 1.3 (DEC 2002) and Gibson et al.
 489 (2011). S = obligate seeder, R = resprouter, Sr = predominantly an obligate seeder, Rs =
 490 predominantly a resprouter. Seed weight is a mean, based on measurement of 30 seeds.

491

<i>Acacia</i> species	Height (m)	Fire response	Seed weight (mg)
<i>A. buxifolia</i>	1 – 4	Sr	20.8
<i>A. caesiella</i>	1 – 3.5	S	19.8
<i>A. cheelii</i>	~7	S	16.1
<i>A. cultriformis</i>	4	S	15.6
<i>A. deanei</i>	1.5 – 7	S	24.7
<i>A. decora</i>	1 – 4	S	14.7
<i>A. doratoxylon</i>	3 – 8	S	9.2
<i>A. gladiiformis</i>	1 – 3	S	21.4
<i>A. implexa</i>	5 – 12	Rs	25.0
<i>A. penninervis</i>	2 – 8	S	72.8

497

498 **Table 3.** Results for Generalised Linear Mixed Models of above ground woody stem density

499 comparing high and low severity sites.

	df	χ^2	P
All species	1	11.552	<0.0001
All <i>Acacia</i> species	1	16.071	<0.0001
<i>Acacia penninervis</i>	1	3.598	0.0578
<i>Acacia cheelii</i>	1	7.237	0.0071

500

501

502 **Figure captions**

503 **Fig. 1** Density of live woody plant stems and all *Acacia* species (mean \pm standard error per m²) in low
504 (■) and high (■) severity sites, and density of *A. cheelii* and *A. penninervis* in high and low severity
505 sites, including only sites where each species was dominant.

506

507 **Fig. 2** Density of individuals (mean \pm standard error per hectare) of each species at the low and high
508 severity sites. Note different y-axis scales on graphs. Density of species is approximately four times
509 higher after high severity fire than after low severity fire

510

511 **Fig. 3** Number of seeds (mean \pm standard error per m²) at two depths in the soil profile, at low or
512 high severity sites subjected to either high or low severity burn. There were significantly more seeds
513 found in low severity sites ($P = 0.0254$), but no significant differences between different depths

514

515 **Fig. 4** Residual seed bank density (mean \pm standard error per m²) of the two dominant *Acacia*
516 species, *A. cheelii* and *A. penninervis* at low (■) and high (■) severity sites

517

518 **Fig. 5** Proportion of soil seed bank samples that contained viable *Acacia* seeds after fire in relation to
519 the number of seedlings within the sample

520

521

- Deleted: Number
- Formatted: Highlight
- Deleted: plant stems
- Formatted: Highlight
- Formatted: Highlight
- Formatted: Highlight

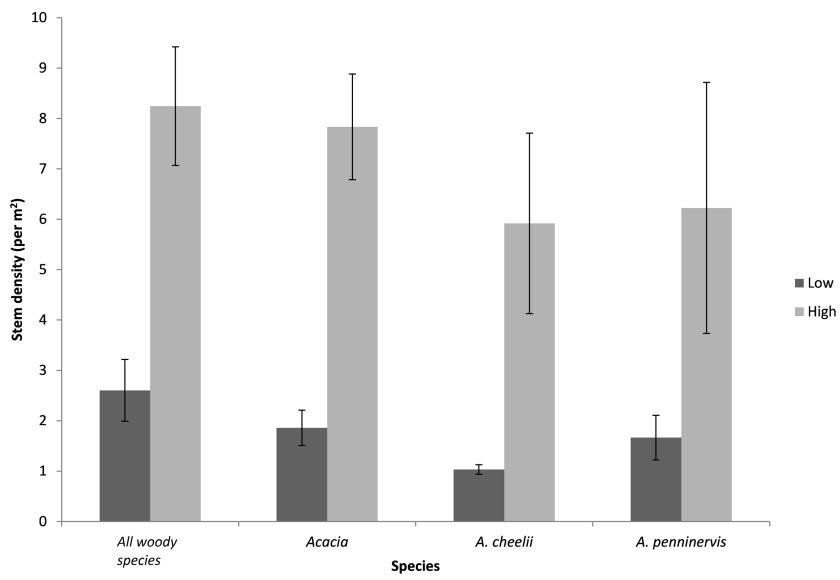
524

525 Figure 1

526

527

528

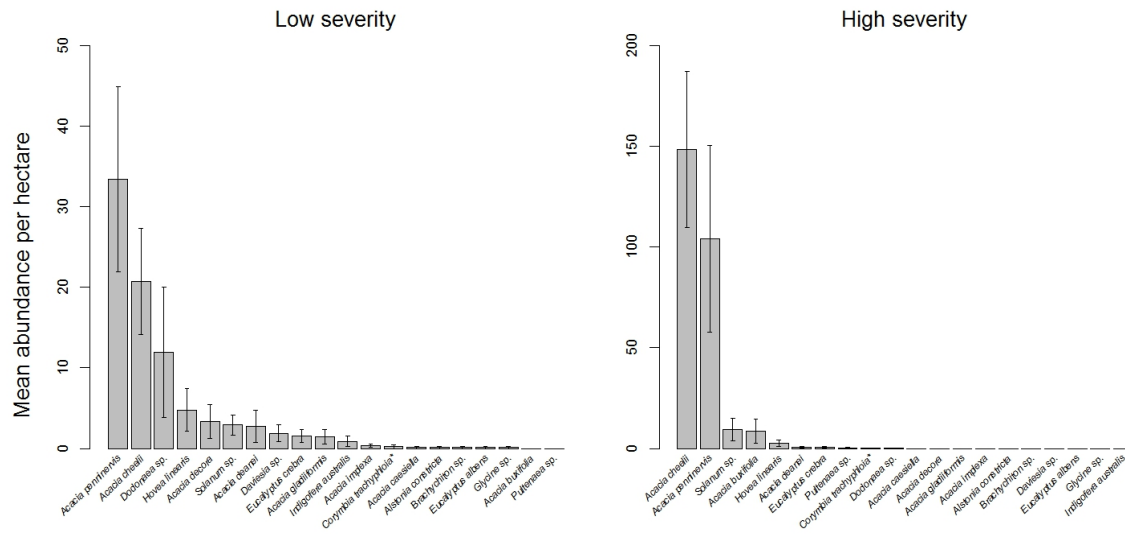


529

530

531 Figure 2

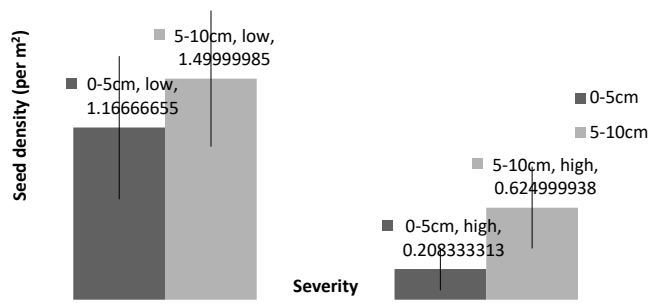
532



533

534 Figure 3

535



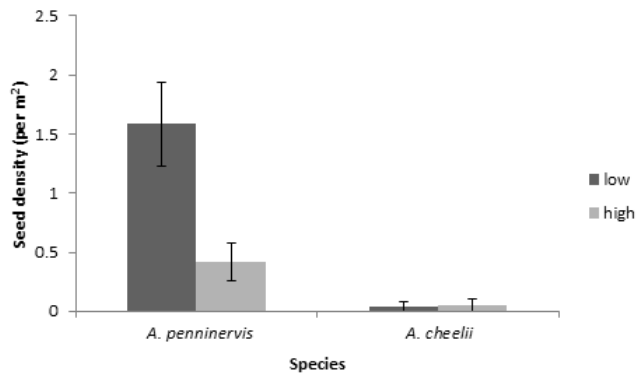
536

537

538

539 Figure 4

540



541

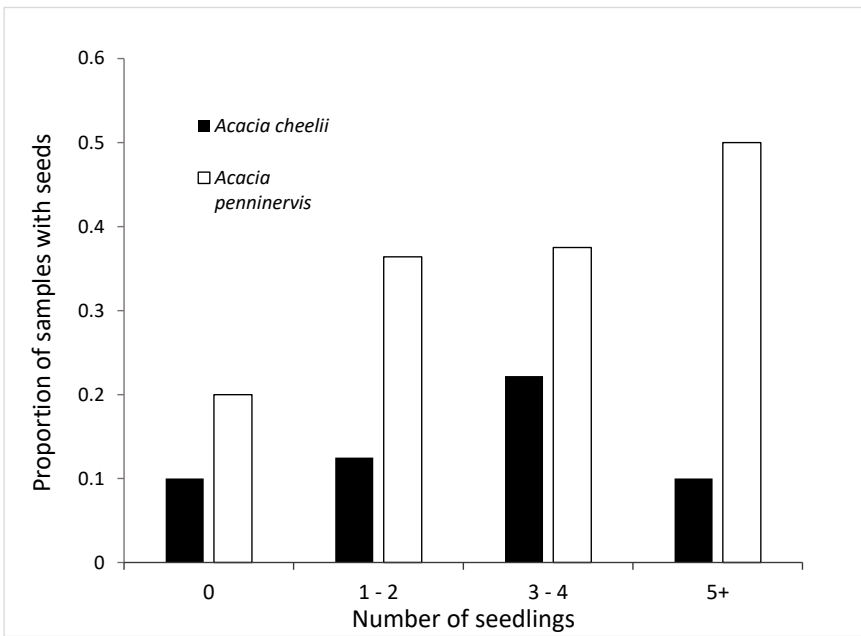
542

543

544 Figure 5

545

546



547

548