



**Biological significance of the dead biomass retention trait in Mediterranean Basin species: an analysis between different successional niches and regeneration strategies as functional groups**

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Review

1 **Biological significance of the dead biomass retention trait in Mediterranean Basin**  
2 **species: an analysis between different successional niches and regeneration**  
3 **strategies as functional groups**

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6 **Running head:** Biological significance of the dead biomass retention trait

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1 **Abstract**

2 Standing dead biomass retention is considered one of the most relevant fuel structural  
3 traits to affect plant flammability. However, very little is known about the biological  
4 significance of this trait and its distribution between different functional groups. Our  
5 aim was to analyse how the proportion of dead biomass produced in Mediterranean  
6 species is related to the successional niche of species (early-, medium- and late-  
7 successional stages) and the regeneration strategy of species (seeders and resprouters).  
8 We evaluated biomass distribution by size classes and standing dead biomass retention  
9 in nine dominant species from the Mediterranean Basin in different development stages  
10 (5, 9, 14 and 26 years since the last fire). The results revealed significant differences in  
11 the standing dead biomass retention of species that presented a distinct successional  
12 niche or regeneration strategy. These differences were restricted to the oldest ages  
13 studied (>9 years). Tree and small tree resprouters, typical in late-successional stages,  
14 presented slight variations with age and a less marked trend to retain dead biomass,  
15 while seeder shrubs and dwarf shrubs, characteristic in early-successional stages,  
16 showed high dead biomass loads. Our results suggest that the species that tend to retain  
17 more dead branches are colonising species that may promote fire in early-successional  
18 stages.

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21 **Keywords:** Dead fuel, Early species, Flammability, Functional traits, Late species,  
22 Resprouter, Seeder.

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## 1 **Introduction**

2 One important point in the ecological understanding of fire regime is to recognise how  
3 biomass traits of plant species can shape it. In fact variation in biomass structure has  
4 referred to changes in the fire regime (Bond & Keeley 2005; Bowman *et al.* 2009), and  
5 classifying biomass according to its flammability is essential for fire hazard  
6 assessments. However, it is extremely difficult to assess plant flammability given the  
7 complex interactions of the components that drive it (Cornelissen *et al.* 2003). Most  
8 approaches have centred on understanding the role of biomass moisture in flammability  
9 (Gill *et al.* 1978; Dimitrakopoulos & Papaioannou 2001), especially how seasonal  
10 variations in live biomass moisture relate with the ignition delay time (Pellizaro *et al.*  
11 2007). Other studies have used discrete biomass elements (e.g., leaves and twigs) for  
12 assessing small-scale intrinsic properties on flammability, such as specific gravity,  
13 mineral content and chemical composition (Fernandes & Cruz 2012). However, very  
14 few studies on the structural distribution of canopy components according to size are  
15 available (Cowan & Ackerly 2010; Saura-Mas *et al.* 2010), particularly on the degree of  
16 standing dead biomass retention (branches and leaves) in different species (see Keeley  
17 *et al.* 2012). A key role in fire ignitions has been attributed to this biomass type  
18 (Santana & Marrs 2014), and it has the peculiarity of being dynamic in both plant  
19 ontogeny and the progression of plant succession (Baeza *et al.* 2011; Santana *et al.*  
20 2013). Therefore, in order to gain a better understanding of the relationship of biomass  
21 structural traits associated with flammability and fire hazard, a better description of  
22 variations in dead and live biomass with time is considered essential.

23 Standing dead biomass retention in the canopy is one of the most relevant plant  
24 properties involved in vegetation flammability (Schwilk & Ackerly 2001; Keeley *et al.*  
25 2012). The relationship between dead and living biomass is involved in key fire

1 occurrence processes as it influences both fire initiation (ignitability) and fire extinction  
2 (Santana & Marrs 2014). Dead biomass is often characterised by its low water content,  
3 which makes it the best point for fire ignition, and the first heat source when fire  
4 commences. Living biomass may either contribute actively to fire energy or serve as a  
5 heat sink to reduce fire intensity and propagation (Pyne *et al.* 1996). In ecosystems  
6 dominated by shrubs, fire behaviour can depend mainly on the proportion of standing  
7 dead biomass present in vegetation (Baeza *et al.* 2002; Anderson & Anderson 2009).  
8 The factors that affect biomass moisture also vary between dead and live biomass. Dead  
9 biomass moisture is controlled mainly by physical processes and depends clearly on  
10 atmospheric conditions. In contrast, live biomass moisture is affected by not only  
11 meteorological factors and soil water availability, but also by the physiological plant  
12 mechanisms that regulate water content (Castro *et al.* 2003). Therefore, the inherent low  
13 water content of standing dead biomass can prove critical during dry seasons and can,  
14 thus, considerably enhance plant flammability.

15 Plant communities may be more or less fire prone depending on the individual  
16 attributes of the species composing them (Fernandes 2009; Keeley *et al.* 2012). Some  
17 species possess structural (canopy architecture) and physiological (water status) traits  
18 (Castro *et al.* 2003), which make them more flammable than others (Baeza *et al.* 2011;  
19 Santana *et al.* 2011). Differences in species flammability with distinct post-fire  
20 regeneration strategies can be key to establish a mutual link between fire regime and  
21 vegetation communities. Recent studies done in Mediterranean-type ecosystems have  
22 found a higher dead branch proportion in species with post-fire seeding as compared to  
23 resprouter species (Saura-Mas *et al.* 2010; Cowan & Ackerly 2010). This fact suggests  
24 that fire is an important selective pressure in the evolution of the post-fire seeding trait  
25 (high post-fire germination) (Keeley *et al.* 2011; Bradshaw *et al.* 2011; He *et al.* 2011;

1 Verdú & Pausas 2013), which benefits those species that promote fire ignition and fire  
2 intensity through dead biomass retention (Schwilk 2003; Keeley *et al.* 2012).  
3 Nevertheless, very few hypotheses have been posed on the selective value of this trait  
4 (high dead biomass retention) other than its effects on flammability (Schwilk 2003;  
5 Keeley *et al.*, 2011; Keeley *et al.* 2012).

6 One point that has been the object of very few studies on the flammability  
7 properties of vegetation is processes that assemble the community structure. For  
8 example, the post-fire successional gradient may lead to changes in resources  
9 availability and, thus, in species composition with different flammability traits, which  
10 could affect the fire regime (Cramer *et al.* 2008; Odion *et al.* 2010). Early-successional  
11 species typically have a set of associated traits, including short life span, and fast-  
12 growing, shade-intolerant, dry-fruited, small-seeded, seed dormancy and high seedling  
13 establishment rates after disturbance (Rees *et al.* 2001). In contrast, late-successional  
14 species show the opposite syndrome (Verdú 2000; Ackerly 2004) and, if no  
15 disturbances occur, they exclude colonizer species as they reduce resources availability  
16 to levels below those they actually require (Huston & Smith 1987; Grubb 1998).  
17 Despite available confirmed information that describes opposite traits between early-  
18 and late-successional species, there is very little information available about  
19 flammability properties, such as biomass size distribution and dead biomass retention  
20 patterns. This point is fundamental for understanding plant flammability patterns during  
21 a successional process over time and their possible effect on the fire regime.

22 In this paper, we assessed biomass structural properties in the canopy of nine  
23 dominant species from the Mediterranean Basin. We measured biomass distribution by  
24 size classes and standing dead biomass retention over a chronosequence of 5-26 years  
25 since the last fire. We tested two initial hypotheses in this paper: 1) the proportion of

1 dead biomass produced in Mediterranean species is related to the successional niche  
2 (early-, medium- and late-successional stage); 2) the proportion of dead biomass is  
3 related to species' regeneration strategies (seeders and resprouters). These two  
4 functional type classifications have already been related to each other in Mediterranean  
5 Basin species (Verdú 2000; Saura-Mas 2008; Verdú *et al.* 2009) and are expected to  
6 share similar traits; i.e., early-successional with seeder species and late-successional  
7 with resprouter species.

## 9 **Materials and Methods**

### 10 *Study site and experimental design*

11 The study area is located in the province of Valencia (hereafter referred to as Ayora) in  
12 the eastern Iberian Peninsula (39°05' - 40°5'N, 0°51' - 1°59'W). Climate is sub-dry meso-  
13 Mediterranean with mean temperatures of 13-17°C and annual mean rainfall of 400-700  
14 mm. Leptosols, stony and shallow soils, are formed on limestone and dolomite  
15 dominates on the uplands of the study area, while regosols, less stony and deeper soils  
16 that form on marl, dominate lower colluvial areas. The landscape is dominated mainly  
17 by shrublands, which have regenerated after fire and have different species  
18 compositions. Dominant shrub species include *Rosmarinus officinalis*, *Ulex parviflorus*  
19 and *Quercus coccifera*. Small patches of trees (i.e., *Quercus ilex* and *Pinus halepensis*)  
20 are found, but no forests. In fact the potential vegetation is considered a sclerophyllous  
21 oak forest of *Quercus ilex*. Based on fire history records of the Regional Valencian  
22 Government's Forest Service, we selected four areas at Ayora which had burned in the  
23 summers of 1979, 1991, 1996 and 2000 (30,000, 5,000, 600 and 50 ha, respectively).  
24 This provided us with a stand-age chronosequence of 26, 14, 9, and 5 years,  
25 respectively, since the last fire. Our vegetation analysis assumed temporal variations

1 within the study area, which is a widely used approach in vegetation dynamics research  
2 to determine different vegetation development stages with time.

3

#### 4 *Sampling*

5 In order to well represent the species in each burnt area (Cornelissen *et al.* 2003), we  
6 selected nine woody species, which represent 85-90% of the vegetation cover at Ayora  
7 (Baeza *et al.* 2007), these being: *Cistus albidus* L., *Cistus clusii* L., *Erica multiflora* L.,  
8 *Juniperus oxycedrus* L., *Pinus halepensis* Miller, *Quercus coccifera* L., *Quercus ilex* L.,  
9 *Rosmarinus officinalis* L. and *Ulex parviflorus* Pour. (nomenclature follows De Bolòs *et*  
10 *al.* 1993). These species included shrubs, dwarf shrubs and trees, early- and late-  
11 successional species, and different post-fire regeneration strategies (seeders and  
12 resprouters) (Table 1).

13 In each fire area, a plot of 2-3 ha was sampled where 10-12 individuals of each  
14 species, separated by at least 5 metres, were randomly selected (375 individuals) and  
15 harvested during vegetation inactive periods to avoid seasonal maximum dead biomass  
16 accumulation in summer (the winters of 2004 and 2005). Any individuals that were  
17 isolated, dead, broken or growing in especially favourable locations were avoided. For  
18 resprouter species, the ramet was considered; i.e., the unsuppressed mid-developed  
19 shoot that sprouts directly from burnt stumps. The age of all the individuals per plot was  
20 checked by counting growth rings in *J. oxycedrus* and *P. halepensis* as being  
21 representative of resprouter and seeder species, respectively (Baeza *et al.* 2011).

22 At each site, the biomass structural variables were measured by species and  
23 fraction (i.e., live and dead biomass, and different diameter classes of biomass). For  
24 each individual plant, biomass was separated into live and dead, and was then grouped  
25 according to diameter class < 6 mm (twigs), 6-25 mm (branches), 25-75 mm (thick



1 branches) and > 75 mm (trunks) (Cornelissen *et al.* 2003). No dead leaf material was  
2 encountered or its contribution to total individual biomass was imperceptible (<1 %).  
3 Live leaves were separated (or phylloides for *U. parviflorus*) and were included in the  
4 live twigs fraction (< 6 mm). Tree trunks (> 75 mm) were not considered to contribute  
5 to fire-consumed biomass. Separated material was oven-dried (3 days at 60°C) to  
6 ascertain its dry weight.

7

### 8 *Statistical analysis*

9 Species were ordinated on the basis of their biomass' structural composition by a  
10 principal component analysis (PCA). Species with a similar biomass structure should be  
11 grouped near each other in the ordination space, which should help identify any  
12 architectural pattern in biomass classes. The analysed data included all the biomass  
13 diameter classes, and redundant variables were avoided; thus the dead and live fractions  
14 of both twigs and branches, and of live thick branches, were used. The biomass data,  
15 grouped according to size class and to dead and live biomass, were transformed into  
16 proportions of each individual's total weight in order to remove large-scale differences  
17 among the original variables. A multivariate analysis was conducted using CANOCO  
18 4.5 for Windows (Ter Braak & Šmilauer 1998).

19 Linear Mixed Models (LMMs) were used to test the two hypotheses presented  
20 herein: (1) the proportion of dead biomass produced in Mediterranean species is related  
21 to the successional niche; (2) the proportion of dead biomass is related to species'  
22 regeneration strategies. In the model construction, we considered the successional niche  
23 (early-, medium- and late-successional stages) as fixed effects for Hypothesis 1,  
24 whereas regeneration strategy (seeders and resprouters) was considered for Hypothesis  
25 2. In order to test the effect of individual age on the dead biomass proportion and its

1 subsequent effect on our hypotheses, ages of individuals were also considered a  
2 continuous fixed effect, together with its interaction with either successional niche or  
3 regeneration strategy. The species nested within successional niche or regeneration  
4 strategy were included as random factors. For these analyses, the proportion of dead  
5 biomass in the percentage was previously arcsine- ( $\sin^{-1} \sqrt{(x/100)}$ ) transformed. LMMs  
6 were performed using the lme4 package and the lmer function in the R software  
7 environment (v.2.12.2: R Development Core Team 2011). The ANOVA function in the  
8 lmerTest package was used to test the global differences between the different  
9 successional niches and regeneration strategies by considering all ages together. Finally,  
10 differences between successional niches and regeneration strategies for each specific  
11 age were analysed by contrasting the least-squares means with the “lsmeans” function.  
12 For this purpose, LMMs were fitted using age as the discrete variable.

13

## 14 **Results**

15 At early ages (5 years), species are composed mainly of live twigs (Fig. 1; see also  
16 Table S1 A-C in the Supplementary Material (SM) for the values obtained for each  
17 species). With time, all the size classes of the live and dead biomasses tended to  
18 increase (except for *C. clusii*), but distinctly between species (SM Table S1 A-C). At  
19 later ages (9-26 years), some shrub species (*C. clusii*, *C. albidus*, *U. parviflorus* and *E.*  
20 *multiflora*) tended to present a very high proportion of dead twigs. These species were  
21 ordinated on the right-hand side of the PCA Axis 1 (Fig. 1). The PCA Axis 2 clearly  
22 discriminated the species in which live thick branch fractions predominated. Two shrub  
23 species (*Q. coccifera* and *R. officinalis*) presented reduced dead twig accumulation,  
24 which were ordinated on the positive part of this axis (Fig. 1). Tree species (*Q. ilex*, *J.*

1 *oxycedrus* and *P. halepensis*) were also located on the positive side of this axis and at  
2 older ages (Fig. 1).

3 The proportion of dead biomass was strongly affected by the age of individuals  
4 in the most functional groups. Initially, there were no significant differences between  
5 successional niches when analysing all the individuals of all the different ages together  
6 (5, 9, 14 and 26 years) (ANOVA; F-value = 0.539, p= 0.606). However, the significant  
7 interaction of successional niches with the age in the fitted LMM (Table 2) suggested  
8 differences at specific ages. In fact when compared for each specific age, no significant  
9 differences were found in the young stages (5 years), when dead biomass accumulation  
10 was still low for all the species (Figure 2A). However, significant differences were  
11 found at older ages (>9 years) between early-successional species and the other  
12 successional niches (medium and late-successional) (Figure 2A). Similarly, no  
13 differences were observed between resprouter and seeder species when analysing the  
14 regeneration strategy for all ages together (ANOVA; F-value = 0.217, P=0.654).  
15 However, a robust interaction with age was observed (Table 2), and significant  
16 differences in accumulated dead biomass at older ages (>9 years) were found between  
17 the two regeneration strategies (Fig. 2B).

18

## 19 **Discussion**

20 The present study reveals considerable differences in the standing dead biomass  
21 retention of the species that presented a distinct successional niche or regeneration  
22 strategy. This is, therefore, in line with our two initial hypotheses. Nonetheless, these  
23 two functional groups showed marked interactions with age, and no differences were  
24 found in young stages given the low proportion of standing dead biomass that  
25 accumulated in all the species. It was at older ages (>9 years) when (i) seeders showed

1 greater dead biomass accumulations than resprouters, and (ii) early-successional species  
2 accumulated more than medium- and late-successional species. Our approach suggests  
3 that the differences in biomass structural traits, and therefore fire hazard, may be  
4 captured by functional groups; i.e. changes in the relative abundance of the different  
5 functional groups may promote changes in flammability at the community level (Bond  
6 & Keeley 2005; Odion *et al.* 2010).

7 It is worth noting that in Mediterranean Basin species, the two functional type  
8 classifications studied herein have already been related to each other (Verdú 2000;  
9 Saura-Mas 2008; Verdú *et al.* 2009) and they are expected to share similar traits; i.e.,  
10 early-successional with seeder species and late-successional with resprouter species.  
11 The two tested hypotheses are, therefore, not completely independent of each other.  
12 However, these results suggest that a new hypothesis about the biological significance  
13 of dead biomass accumulation, which can be associated with the species' successional  
14 niche. To date, these differences were tested only between different regeneration  
15 strategies and at a fixed point with time (Cowan and Ackerly 2010; Saura-Mas *et al.*  
16 2010).

17 Different structural compositions in both condition types (live and dead, Fig. 1)  
18 and diameter classes were found among the studied species. These patterns also shifted  
19 between functional groups. At early ages (5 years), young biomass dominated in all the  
20 species with high proportions of live twigs. In these stages, all the species were less  
21 flammable and wildfires could progress only under highly flammable conditions  
22 (extreme fire weather) (Moritz *et al.* 2004). Yet the results we obtained in the  
23 intermediate and old stages revealed two different patterns: a group of early-  
24 successional dwarf shrubs and shrubs, mainly seeders (*C. albidus*, *C. clusii*, *E.*  
25 *multiflora* and *U. parviflorus*), with a high proportion of dead twigs and dead branches;

1 another group of bigger species (small trees and trees such as *Q. coccifera*, especially *J.*  
2 *oxycedrus* and *Q. ilex*), which are typical of late-successional stages and mainly  
3 resprouters. This second group presented a higher proportion of live thick branches and  
4 lesser standing dead biomass retention. Therefore based on structural biomass  
5 organisation, our results suggested that shrubs, especially dwarf shrubs, exhibit a more  
6 flammable canopy architecture, which is dominated by the fine biomass class and a high  
7 proportion of dead biomass. In contrast, trees and small trees are not as flammable, tend  
8 to accumulate less standing dead biomass and have a poorer canopy structure dominated  
9 by a coarse biomass class. These findings are consistent with the positive feedback  
10 obtained between shrublands and fire (Díaz-Delgado *et al.* 2002; Syphard *et al.* 2006),  
11 while forests and woodlands are less wildfire-prone (Bond *et al.* 2005; Mermoz *et al.*  
12 2005; Odion *et al.* 2010). The lower accumulation of dead biomass in early-  
13 successional species may also suggest a greater likelihood of burning all year round,  
14 whereas the opportunities to burn in late-successional species would be more limited to  
15 specific seasons. Live foliage moisture decreases under flammable levels only as  
16 summer progresses, which is when species poor in dead biomass have more similar  
17 burning characteristics to species with rich dead biomass. Nonetheless, it should be  
18 taken into account that to draw these conclusions, we only considered the aerial  
19 structure of biomass because in some cases species whose canopy does not retain dead  
20 foliage can, in turn, accumulate flammable litter underneath (Scarff & Westoby 2006).  
21 For example, Curt *et al.* (2011) observed in Mediterranean ecosystems in south-east  
22 France that the litter that accumulated in early-successional stages (dominated by *Cistus*  
23 sp.) was less flammable than the litter from late-successional stages dominated by  
24 *Quercus* sp.

1           Despite the retention of dead branches being one of the most important plant  
2 properties to enhance plant flammability (Schwilk & Ackerly 2001; Keeley *et al.* 2012),  
3 information on the relation to this property and its possible biological significance is  
4 scarce. In Mediterranean-type ecosystems, it has been previously hypothesised that the  
5 evolution of post-fire regenerations traits coincides with the natural selection of  
6 enhancing flammability traits, such as retention of dead branches through increased  
7 post-fire recruitment (Schwilk & Ackerly 2001; He *et al.* 2011; Keeley *et al.* 2011;  
8 Pausas *et al.* 2012; Keeley *et al.* 2012). Dead biomass may enhance species  
9 flammability with germination stimulated by heat of fire, along with a later flush in  
10 recruitment. In fact it has been observed that the species which retain dead branches  
11 produce higher temperatures and longer residence times in soil during combustion  
12 (Santana *et al.* 2011). This suggests a link between post-fire regenerative strategy and  
13 flammability traits, which may result in a fitness advantage for more flammable species.  
14 Seeder species with fire-enhanced recruitment (i.e., *Adenostoma*, *Ceanothus*, *Ulex* or  
15 *Cistus*) display a marked tendency to retain dead branches in the canopy (Schwilk 2003;  
16 Saura-Mas *et al.* 2010). In contrast, those resprouter species capable of recruiting when  
17 fire is absent, such as *Quercus*, *Prunus* and *Rhamnus* species, are expected to have a  
18 lower standing biomass fuel proportion (Keeley *et al.* 2012). However, previous studies  
19 have also shown that post-fire recruitment in some species is not completely dependent  
20 on heat exposure and has, therefore, called into question the flammability selection  
21 hypothesis for enhancing post-fire recruitment. For instance, daily temperature  
22 fluctuation during summer periods may have a stronger influence during the  
23 germination of heat-stimulated species with considerable dead biomass accumulations  
24 (Baeza & Roy 2008; Santana *et al.* 2013a; Ooi *et al.* 2014). This suggests that in  
25 flammable species which retain dead branches, for example *Ulex parviflorus*, not only

1 the heat generated by fire can break seed dormancy, but also other factors such as soil  
2 temperature fluctuations are also efficient germination triggers.

3 Pausas *et al.* (2012) observed that the quantity of dead biomass in *U. parviflorus*  
4 for high fire recurrence scenarios was no larger than in low fire occurrence scenarios.  
5 Other theories have also been posed, such as pyrogenicity as a form of self-protection in  
6 fire-prone environments, where standing dead biomass retention promotes combustion  
7 quickly, should lower local soil heating, and should consequently enhance seed bank  
8 survival (Gagnon *et al.* 2010). Evidently there is still some uncertainty as to the possible  
9 biological significance of why some species accumulate larger quantities of dead  
10 biomass than others.

11 Our results describe a general pattern: those species that tend to retain more dead  
12 branches are pioneer species of early-successional stages after land abandonment, or  
13 when vegetation is removed by fire and other disturbances (Baeza *et al.* 2007; Baeza *et*  
14 *al.* 2011), and they are often obligate seeder species (Verdú 2000; Lloret *et al.* 2005).  
15 This finding may suggest a new dead biomass retention hypothesis linked to the  
16 successional niche of species. Early-successional species often show high seedling  
17 establishment rates, quick attainment of reproductive age, rapid growth, fast turnover of  
18 organs (leaves), short life span and shade intolerance (Bazzaz 1979; Rees *et al.* 2001;  
19 Ackerly 2004). These species are considered competitive in environments with high  
20 resource availability, such as open spaces, (Rees *et al.* 2001). Early-successional species  
21 are expected to display prolific branching, which could reduce light availability and, in  
22 certain cases, could even increase the dead wood component of the lowest branches  
23 when they are considerable in size and of a mature age (Schlesinger & Gill 1980). In  
24 fact it has been observed that many Mediterranean early-successional species do not  
25 possess an effective self-pruning mechanism, but accumulate dead branches on the

1 canopy, whereas late-successional species, like *Arbutus unedo* and some *Quercus* sp.,  
2 display effective self-pruning, which concentrates mainly in autumn (Montserrat-Martí  
3 *et al.* 2004). Differences in resource allocation patterns between shoots and roots have  
4 also been suggested to be a possible determinant of dead biomass accumulation (Cowan  
5 & Ackerly 2010). Species with a high shoot:root ratio are expected to be less drought-  
6 tolerant in summer, hence the dieback in part of the biomass might increase during  
7 summer droughts (Montserrat-Martí *et al.* 2004). This pattern has also been noted in the  
8 Mediterranean Basin where early-successional species, such as *U. parviflorus* and *R.*  
9 *officinalis*, show a higher shoot:root ratio than others like *Pistacia lentiscus* or *Myrtus*  
10 *communis*, which are typical of more advanced successional stages (Hernandez *et al.*  
11 2010; Saura-Mas & Lloret 2013).

12 In summary, our study stresses that the ability to retain dead branches in the  
13 canopy may be the result of successional processes in response to resource availability  
14 along the successional gradient. Early-successional communities, which have adapted to  
15 respond to an environment with high seasonal variability, are dominated by both shrub  
16 and dwarf shrub obligate seeders, these being drought-tolerant species with semi-  
17 deciduous leaves that help regulate the evapotranspiration surface in summer (Saura-  
18 Mas & Lloret 2007). However, these species have a poor branch self-pruning capacity  
19 that enhances flammability. In contrast, late-successional communities are dominated  
20 by drought-avoiding species, which include evergreen sclerophyllous trees and small  
21 trees resprouters with deep-rooted systems. These species possess an efficient self-  
22 pruning mechanism and are, consequently, less flammable.

23



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7

## 8 **References**

9

- 10 Ackerly D.D. (2004) Functional strategies of chaparral shrubs in relation to seasonal  
11 water deficit and disturbance. *Ecological Monographs* **74**, 25–44.
- 12 Anderson S.A.J., Anderson W.R. (2009) Predicting the elevated dead fine fuel moisture  
13 content in gorse (*Ulex europaeus* L.) shrub fuels. *Canadian Journal of Forest*  
14 *Research* **39**, 2355-2368.
- 15 Baeza M.J., Roy J. (2008) Germination of an obligate seeder (*Ulex parviflorus*) and  
16 consequences for wildfire management. *Forest Ecology and Management* **256**,  
17 685–693.
- 18 Baeza M.J., De Luís M., Raventós J., Escarré A. (2002) Factors influencing fire  
19 behaviour in shrublands of different stand ages and the implications for using  
20 prescribed burning to reduce wildfire risk. *Journal of Environmental*  
21 *Management* **65**, 199-208.
- 22 Baeza M.J., Valdecantos A., Alloza J.A., Vallejo V.R. (2007) Human disturbance and  
23 environmental factors as drivers of long-term post-fire regeneration patterns in  
24 Mediterranean forest. *Journal of Vegetation Science* **18**, 243-252.

- 1 Baeza M.J., Santana V.M., Pausas J.G., Vallejo V.R. (2011) Successional trends in  
2 standing dead biomass in Mediterranean basin species. *Journal of Vegetation*  
3 *Science* **22**, 467-474.
- 4 Bazzaz F.A. (1979) The physiological ecology of plant succession. *Annual Review of*  
5 *Ecological Systems* **10**, 351-371.
- 6 Bond W.J., Keeley J.E. (2005) Fire as a global ‘herbivore’: the ecology and evolution of  
7 flammable ecosystems. *Trends in Ecology and Evolution* **20**, 387-394.
- 8 Bond W.J., Woodward F.I., Midgley G.F. (2005) The global distribution of ecosystems  
9 in a world without fire. *New Phytologist* **165**, 525-538.
- 10 Bowman D.M.J.S., Balch J.K., Artaxa P., Bond W.J., Carlson J.M., Cochrane M.A.,  
11 D’Antonio C.M., DeFries R.S., Doyle J.C., Harrison S.P., Johnston F.H., Keeley  
12 J.E., Krawchuk M.A., Kull C.A., Marston J.B., Moritz M.A., Prentice I.C., Roos  
13 C.I., Scott A.C., Swetnam T.W., van der Werf G.R., Pyne S.J. (2009) Fire in the  
14 earth system. *Science* **324**, 481–484.
- 15 Bradshaw S. D., Dixon K.W., Hopper S.D., Lambers H., Turner S.R. (2011) Response  
16 to Keeley *et al.*: Fire as an evolutionary pressure shaping plant traits. *Trends in*  
17 *Plant Science* **16**, 405.
- 18 Castro F.X., Tudela A., Sebastià M.T. (2003) Modeling moisture content in shrubs to  
19 predict fire risk in Catalonia (Spain). *Agricultural and Forest Meteorology* **116**,  
20 49–59.
- 21 Cornelissen J.H., Lavorel S., Garnier E., Díaz S., Buchmann N., Gurvich D.E., Reich  
22 P.B., ter Steege H., Morgan H.D., van der Heijden M.G.A., Pausas J.G., Poorter  
23 H. (2003) A handbook of protocols for standardised and easy measurement of  
24 plant functional traits worldwide. *Australian Journal of Botany* **51**, 335–380.

- 1 Cowan P., Ackerly D. (2010) Post-fire regeneration strategies and flammability traits of  
2 California chaparral shrubs. *International Journal of Wildland Fire* **19**, 984–989.
- 3 Cramer A., Hobbs R.J., Standish R.J. (2008) What's new about old fields? Land  
4 abandonment and ecosystem assembly. *Trends in Ecology and Evolution* **23**,  
5 104-112.
- 6 Curt T., Schaffhauser A., Borgniet L., Dumas C., Estève R., Gantaume A., Jappiot M.,  
7 Martin W., N'Diaye A., Poilvet B. (2011) Litter flammability in oak woodlands  
8 and shrublands of southeastern France. *Forest Ecology and Management* **261**,  
9 2214-2222.
- 10 De Bolòs O., Vigo J., Masalles R.M., Ninot J.M. (1993) Flora Manual dels Països  
11 Catalans. 2nd. Ed. Pòrtic, Barcelona. 1233 pp.
- 12 Díaz-Delgado R., Lloret F., Pons X., Terradas J. (2002) Satellite evidence of decreasing  
13 resilience in Mediterranean plant communities after recurrent wildfire. *Ecology*  
14 **83**, 2293-2303.
- 15 Dimitrakopoulos A.P., Papaioannou K.K. (2001) Flammability assessment of  
16 Mediterranean forest fuels. *Fire Technology* **37**, 143-152.
- 17 Fernandes P. M. (2009). Combining forest structure data and fuel modelling to assess  
18 fire hazard in Portugal. *Annals of Forest Science* **66**, 4. 415p1-415p9.
- 19 Fernandes P. M., Cruz, M. G. (2012). Plant flammability experiments offer limited  
20 insight into vegetation–fire dynamics interactions. *New Phytologist* **194** (3), 606-  
21 609.
- 22 Gagnon P.R., Passmore H.A., Platt W.J., Myers J.A., Paine T., Harms K.E. (2010) Does  
23 pyrogenicity protect burning plants. *Ecology* **91**, 3481-3486.
- 24 Gill A.M., Trollope W.S., MacArthur D.A. (1978) Role of moisture in the flammability  
25 of natural fuels in the laboratory. *Australian Forest Research* **8**, 199-208.

- 1 Grubb P.J. (1998) A reassessment of the strategies of plants which cope with shortages  
2 of resources. *Perspectives in Plant Ecology, Evolution and Systematics* 1, 3-31.
- 3 He T., Lamont B.B., Downes K.S. (2011) Banksia born to burn. *New Phytologist* **191**,  
4 184–196.
- 5 Hernández E.I., Vilagrosa A., Pausas J.G., Bellot J. (2010) Morphological traits and  
6 water use strategies in seedlings of Mediterranean coexisting species. *Plant*  
7 *Ecology* **207**, 233-244.
- 8 Huston M., Smith T. (1987) Plant succession: life history and competition. *American*  
9 *Naturalist* **130** (2), 169-198.
- 10 Keeley J.E., Pausas J.G., Rundel P.W., Bond W.J., Bradstock R. A. (2011) Fire as an  
11 evolutionary pressure shaping plant traits. *Trends in Plant Science* **16**, 406–411.
- 12 Keeley J.E., Bond W.J., Bradstock R.A., Pausas J.G., Rundel P.W. (2012) Fire in  
13 Mediterranean Ecosystems: Ecology, Evolution and Management. (Cambridge  
14 University Press. New York). 515 pp.
- 15 Lloret F., Estevan H., Vayreda J., Terradas J. (2005) Fire regenerative syndromes of  
16 forest woody species across fire and climatic gradients. *Oecologia* **146**, 461-468.
- 17 Mermoz M., Kitzberger T., Veblen T.T. (2005) Landscape influences on occurrence and  
18 spread of wildfires in Patagonian forests and shrublands. *Ecology* **86**, 2705-  
19 2715.
- 20 Monserrat Martí G., Palacio S., Milla R. (2004) Fenología y características funcionales  
21 de las plantas leñosas mediterráneas. In '*Ecología del bosque mediterráneo en*  
22 *un mundo cambiante*'. (Ed Valladares F.). (Ministerio de Medio Ambiente.  
23 EGRAF SA. Madrid). pp. 129-162.

- 1 Moritz, M.A., Keeley J.E., Johnson E.A., Schaffner A.A. (2004). Testing a basic  
2 assumption of shrubland fire management: How important is fuel age? *Frontiers*  
3 *in Ecology and the Environment* **2**, 67-72.
- 4 Odion D.C., Moritz M.A., DellaSala D.A. (2010) Alternative community states  
5 maintained by fire in the Klamath Mountains. USA. *Journal of Ecology* **98**, 96-  
6 105.
- 7 Ooi M. K., Denham, A. J., Santana, V. M., Auld, T. D. (2014) Temperature thresholds  
8 of physically dormant seeds and plant functional response to fire: variation  
9 among species and relative impact of climate change. *Ecology and Evolution* **4**  
10 (5), 656-671.
- 11 Paula S., Arianoutsou M., Kazanis D., Tavsanoğlu Ç., Lloret F., Buhk C., Ojeda F.,  
12 Luna B., Moreno J., Rodrigo A., Espelta J., Palacio S., Fernández-Santos B.,  
13 Fernandes P., Pausas J.G. (2009) Fire-related traits for plant species of the  
14 Mediterranean Basin. *Ecology* **90**, 1420.
- 15 Pausas J.G., Alessio G., Moreira B., Corcobado G. (2012) Fires enhance flammability  
16 in *Ulex parviflorus*. *New Phytologist* **193**, 18–23.
- 17 Pellizzaro G., Duce P., Ventura A., Zara P. (2007) Seasonal variations of live moisture  
18 content and ignitability in shrubs of the Mediterranean basin. *International*  
19 *Journal of Wildland Fire* **16**, 633-641.
- 20 Pyne S.J., Andrews P.J., Laven R.D. (Eds) (1996) 'Introduction to wildland fire'.  
21 Second edition (John Wiley & Sons New York-Chichester UK). 769 pp.
- 22 Rees M., Condit R., Crawley M., Pacala S., Tilman D. (2001) Long-term studies of  
23 vegetation dynamics. *Science* **293**, 650-655.

- 1 Santana V.M., Baeza M.J., Vallejo V.R. (2011) Fuel structural traits modulating soil  
2 temperatures in different species patches of Mediterranean Basin shrublands.  
3 *International Journal of Wildland Fire* **20**, 668-677.
- 4 Santana V. M, Baeza M. J., Marrs R.H. (2013) Response of woody and herbaceous fuel  
5 to repeated fires in Mediterranean gorse shrublands. *International Journal of*  
6 *Wildland Fire* **22**, 508-514.
- 7 Santana V.M., Baeza M.J., Blanes M.C. (2013a) Clarifying the role of heat of fire and  
8 daily temperature fluctuation as germination cues for Mediterranean Basin  
9 obligate seeders. *Annals of Botany* **111**, 127-134.
- 10 Santana V.M., Marrs R.H. (2014) Flammability properties of British heathland and  
11 moorland vegetation: Models for predicting fire ignition. *Journal of*  
12 *Environmental Management* **139**, 88-96.
- 13 Saura-Mas S. (2008) Caracterització funcional d'espècies llenyoses mediterrànies amb  
14 diferents estratègies regeneratives post-incendi. Ph. D. Thesis. Universidad  
15 Autònoma de Barcelona.
- 16 Saura-Mas S., Lloret F. (2007) Adult root structure of Mediterranean shrubs:  
17 relationship with post-fire regenerative syndrome. *Plant Biology* **16**, 147-154.
- 18 Saura-Mas S., Paula S., Pausas J.G., Lloret F. (2010) Fuel loading and flammability in  
19 the Mediterranean Basin woody species with different post-fire regenerative  
20 strategies *International Journal of Wildland Fire* **19**, 783-794.
- 21 Saura-Mas S., Lloret F. (2013) Leaf and shoot water content and leaf dry matter content  
22 of Mediterranean Woody species with different post-fire regenerative strategies.  
23 *Annals of Botany*, **99** (3), 545-554.
- 24 Scarff F.R., Westoby M. (2006). Leaf litter flammability in some semi-arid Australian  
25 woodlands. *Functional Ecology* **20**, 745-752.

- 1 Schlesinger W.H., Gill D. S. (1980) Biomass, production, and changes in the  
2 availability of light, water, and nutrients during the development of pure stands  
3 of the chaparral shrub, *Ceanothus megacarpus* after fire. *Ecology* **61**(4), 781-  
4 789.
- 5 Schwilk D.W. (2003) Flammability is a niche construction trait: canopy architecture  
6 affects fire intensity. *American Naturalist* **162**, 725-733.
- 7 Schwilk D.W., Ackerly D.D. (2001) Flammability and serotiny as strategies: correlated  
8 evolution in pines. *Oikos* **94**, 326-336.
- 9 Syphard A.D., Franklin J., Keeley J.E. (2006) Simulating the effects of frequent fires on  
10 southern California coastal shrublands. *Ecological Applications* **16**, 1744–1756.
- 11 Ter Braak C.J.F., Šmilauer P. (Eds) (1998) 'CANOCO reference manual and users  
12 guide to Canoco for Windows: Software for Canonical community ordination  
13 (version 4). (Microcomputer Power, Ithaca, NY, US).
- 14 Verdú M. (2000) Ecological and evolutionary differences between Mediterranean  
15 seeders and resprouters. *Journal of Vegetation Science* **11**, 265-268.
- 16 Verdú M., Pausas J.G. (2013) Syndrome-driven diversification in a Mediterranean  
17 ecosystem. *Evolution* **67**, 1756-1766.
- 18 Verdú M., Rey P. J., Alcantara J. M., Siles G., Valiente-Banuet A. (2009) Phylogenetic  
19 signatures of facilitation and competition in successional communities. *Journal*  
20 *of Ecology* **97**, 1171-1180.
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2 Tables

3

4 Table 1. Functional traits of the different study species and the successional stage from  
5 the conceptual vegetation dynamics model at Ayora (Baeza *et al.* 2007). Data are taken  
6 from Paula *et al.* (2009) and personal observations (life span and time to maturity).

7

Species	Family	Life form	Regeneration strategy	Life span (years)	Time to maturity (years)	Successional niche
<i>Cistus albidus</i>	Cistaceae	Dwarf shrub	Seeder	Short (10-15)	Short (2)	Early
<i>Cistus clusii</i>	Cistaceae	Dwarf shrub	Seeder	Short (10-15)	Short (2)	Early
<i>Erica multiflora</i>	Ericaceae	Shrub	Resprouter	Medium (30-40)	Short (3-4)	Medium
<i>Juniperus oxycedrus</i>	Cupressaceae	Small tree	Resprouter	Long (>50)	Medium (5-10)	Late
<i>Pinus halepensis</i>	Pinaceae	Tree	Seeder	Very long (> 200)	Long (10-15)	Early
<i>Quercus coccifera</i>	Fagaceae	Shrub	Resprouter	Long (>50)	Medium (5-10)	Late
<i>Quercus ilex</i>	Fagaceae	Tree	Resprouter	Very long (> 200)	Long (15-20)	Late
<i>Rosmarinus officinalis</i>	Lamiaceae	Shrub	Seeder	Medium (30-40)	Short (2-3)	Medium
<i>Ulex parviflorus</i>	Fabaceae	Shrub	Seeder	Short (15-20)	Short (2-3)	Early

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12 Table 2. Parameters estimated by the LMM models for dead fuel retention.

13

Hypothesis	Baseline	Fixed effects	Estimate	SE	t value	P
Successional niche (1)	Early	Intercept	0.061	0.052	1.175	0.279
		Medium	-0.095	0.091	-1.038	0.334
		Late	-0.034	0.081	-0.431	0.679
		Age	0.009	0.001	9.925	<b>&lt;0.001</b>
		Medium x Age	-0.003	0.002	-2.029	<b>0.043</b>
		Late x Age	-0.007	0.001	-5.296	<b>&lt;0.001</b>
Regeneration strategy (2)	Resprouters	Intercept	0.009	0.056	0.17	0.869
		Seeder	0.034	0.075	0.466	0.654
		Age	0.003	0.001	3.204	<b>0.001</b>
		Seeder x Age	0.005	0.001	3.847	<b>&lt;0.001</b>

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1 **Figure legends**

2

3 **Fig. 1.** PCA ordination biplot showing the ordination of species at different stand ages  
4 as compared to the first two PCA axes. The first two axes explain up to 80.6% of  
5 variance in species ordination. Arrows show the structural explanatory variables of live  
6 and dead fuel according to diameter class (twigs, branches and thick branches). PCA  
7 Axis 1 shows a marked negative correlation with live twigs ( $r=0.63$ ;  $P<0.0001$ ) and a  
8 marked positive correlation with dead twigs ( $r=0.33$ ;  $P=0.0003$ ). PCA Axis 2 correlates  
9 positively with thick branches ( $r=0.93$ ;  $P<0.0001$ ). For simplicity, numbers are used to  
10 indicate the first and last stand ages per species (5 and 26 years), but all the ages studied  
11 are shown, following the direction of the arrows (5, 9, 14 and 26 years). C= *C. clusii*,  
12 CA= *C. albidus*, E= *E. multiflora*, J= *J. oxycedrus*, P= *P. halepensis*, QC= *Q. coccifera*,  
13 Q= *Q. ilex*, R= *R. officinalis* and U= *U. parviflorus*.

14

15 **Fig. 2.** Proportion of dead fuel for the different stand ages since the last fire between the  
16 functional groups: A) Successional niches (early-, medium- and late-successional  
17 species); B) Regeneration strategy (seeder and resprouter species). An asterisk denotes  
18 significant differences ( $p< 0.05$ ) for a specific age.

Supplementary material

**Biological significance of the dead biomass retention trait in Mediterranean Basin species: an analysis between different successional niches and regeneration strategy as functional groups**

**Running head:** Biological significance of the dead biomass retention trait

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Supplementary material Table S1. Mean dry weight (g  $\pm$  SE) for live and dead fuel for study species at 5, 9, 14 and 26 years after fire. The results are shown according to size classes: A) Twigs, B) Branches and C) Thick branches. The temporal trend of fuel is indicated in the last column ( $\downarrow$ : Decrease with time,  $\uparrow$ : Increase with time, and  $\Delta$ : Reach the highest values at intermediate stages).

## A): Twigs

Species	Fuel type	5	9	14	26	Tendency
<i>C. albidus</i>	Live	31.6 $\pm$ 7.9	58.7 $\pm$ 10.9	179.3 $\pm$ 26.1	116.2 $\pm$ 23.3	$\Delta$
	Dead	9.2 $\pm$ 2.8	16.9 $\pm$ 5.0	82.4 $\pm$ 14.6	75.1 $\pm$ 12.9	$\Delta$
<i>C. clusii</i>	Live	82.7 $\pm$ 6.4	35.3 $\pm$ 6.1	79.7 $\pm$ 7.8	59.9 $\pm$ 8.6	$\downarrow$
	Dead	3.6 $\pm$ 0.6	1.7 $\pm$ 0.5	22.7 $\pm$ 10.7	26.2 $\pm$ 5.7	$\uparrow$
<i>E. multiflora</i>	Live	183.7 $\pm$ 37.1	490.3 $\pm$ 69.1	554.0 $\pm$ 100.8	388.7 $\pm$ 51.4	$\Delta$
	Dead	1.2 $\pm$ 0.4	9.7 $\pm$ 1.9	56.9 $\pm$ 13.9	134.4 $\pm$ 30.1	$\uparrow$
<i>J. oxycedrus</i>	Live	63.9 $\pm$ 17.7	85.7 $\pm$ 10.6	396.9 $\pm$ 60.9	704.2 $\pm$ 75.1	$\uparrow$
	Dead	0	2.2 $\pm$ 0.6	11.3 $\pm$ 2.5	33.9 $\pm$ 8.0	$\uparrow$
<i>P. halepensis</i>	Live	441.4 $\pm$ 70.6	733.5 $\pm$ 58.5	2540.5 $\pm$ 284.2	18938.1 $\pm$ 1157.9	$\uparrow$
	Dead	0	35.4 $\pm$ 9.4	108.2 $\pm$ 23.6	2489.8 $\pm$ 368.9	$\uparrow$
<i>Q. coccifera</i>	Live	86.7 $\pm$ 13.5	45.0 $\pm$ 9.6	182.8 $\pm$ 52.3	229.3 $\pm$ 35.9	$\uparrow$
	Dead	2.0 $\pm$ 0.7	5.3 $\pm$ 3.0	21.6 $\pm$ 8.9	50.5 $\pm$ 10.9	$\uparrow$
<i>Q. ilex</i>	Live	210.0 $\pm$ 19.8	131.6 $\pm$ 17.0	1148.8 $\pm$ 186.7	1688.1 $\pm$ 201.6	$\uparrow$
	Dead	2.22 $\pm$ 0.7	22.9 $\pm$ 7.5	115.3 $\pm$ 14.9	206.1 $\pm$ 40.7	$\uparrow$
<i>R. officinalis</i>	Live	97.5 $\pm$ 16.7	270.1 $\pm$ 50.6	248.6 $\pm$ 54.9	474.0 $\pm$ 166.9	$\uparrow$
	Dead	0.5 $\pm$ 0.1	3.9 $\pm$ 1.7	14.4 $\pm$ 8.9	51.4 $\pm$ 13.6	$\uparrow$
<i>U. parviflorus</i>	Live	17.3 $\pm$ 4.2	27.8 $\pm$ 5.5	35.9 $\pm$ 7.5	89.3 $\pm$ 16.0	$\uparrow$
	Dead	1.4 $\pm$ 0.3	76.2 $\pm$ 15.1	119.3 $\pm$ 26.0	214.7 $\pm$ 31.6	$\uparrow$

## Table S1 (Continued) B: Branches

Species	Fuel type	5	9	14	26	Tendency
<i>C. albidus</i>	Live	19.7 $\pm$ 5.1	40.4 $\pm$ 7.5	164.8 $\pm$ 28.1	192.1 $\pm$ 38.7	$\uparrow$
	Dead	0	0	14.9 $\pm$ 5.2	43.7 $\pm$ 20.1	$\uparrow$
<i>C. clusii</i>	Live	19.2 $\pm$ 2.1	13.4 $\pm$ 3.1	108.1 $\pm$ 19.7	77.6 $\pm$ 18.5	$\Delta$
	Dead	0	0	0	28.3 $\pm$ 9.3	$\uparrow$
<i>E. multiflora</i>	Live	6.9 $\pm$ 2.0	69.2 $\pm$ 19.3	204.4 $\pm$ 56.8	327.3 $\pm$ 40.9	$\uparrow$
	Dead	0	0	0	26.1 $\pm$ 10.6	$\uparrow$
<i>J. oxycedrus</i>	Live	33.0 $\pm$ 11.3	33.8 $\pm$ 3.6	194.8 $\pm$ 20.7	324.3 $\pm$ 46.5	$\uparrow$
	Dead	3.8 $\pm$ 1.9	0.3 $\pm$ 0.1	41.0 $\pm$ 18.2	71.2 $\pm$ 51.6	$\uparrow$
<i>P. halepensis</i>	Live	130.5 $\pm$ 23.4	309.9 $\pm$ 13.2	1219.2 $\pm$ 187.9	15241.9 $\pm$ 1202.8	$\uparrow$
	Dead	0	30.7 $\pm$ 19.5	195.2 $\pm$ 67.3	4222.5 $\pm$ 552.5	$\uparrow$
<i>Q. coccifera</i>	Live	45.8 $\pm$ 5.8	35.1 $\pm$ 11.3	168.6 $\pm$ 49.9	335.6 $\pm$ 50.7	$\uparrow$
	Dead	4.8 $\pm$ 1.9	0.08 $\pm$ 0.05	14.5 $\pm$ 5.5	15.7 $\pm$ 3.6	$\uparrow$
<i>Q. ilex</i>	Live	154.4 $\pm$ 17.6	187.7 $\pm$ 24.2	1033.2 $\pm$ 139.5	2108.0 $\pm$ 246.0	$\uparrow$
	Dead	0	0	12.2 $\pm$ 7.7	100.1 $\pm$ 29.7	$\uparrow$
<i>R. officinalis</i>	Live	44.6 $\pm$ 9.7	162.6 $\pm$ 25.5	262.5 $\pm$ 51.1	386.7 $\pm$ 99.4	$\uparrow$
	Dead	0	0	0	21.3 $\pm$ 7.8	$\uparrow$
<i>U. parviflorus</i>	Live	0.3 $\pm$ 0.1	31.9 $\pm$ 7.4	124.8 $\pm$ 30.8	327.5 $\pm$ 3.4	$\uparrow$
	Dead	0	0.9 $\pm$ 0.7	10.1 $\pm$ 4.2	176.2 $\pm$ 52.4	$\uparrow$

Table S1 (Continued) C: Thick Branches

Species	Fuel type	5	9	14	26	Tendency
<i>C. albidus</i>	Live	0	0	21.1 ± 8.5	16.0 ± 11.0	Λ
<i>C. clusii</i>	Live	0	0	0	0	
<i>E. multiflora</i>	Live	0	0	0	0	
<i>J. oxycedrus</i>	Live	0	0	142.8 ± 41.7	600.9 ± 72.4	↑
<i>P. halepensis</i>	Live	46.4 ± 14.5	240.3 ± 42.5	1381.6 ± 209.2	8851.8 ± 745.9	↑
<i>Q. coccifera</i>	Live	0	0	0	108.2 ± 59.4	↑
<i>Q. ilex</i>	Live	11.3 ± 7.9	11.2 ± 7.8	1380.2 ± 312.5	3848.9 ± 319.7	↑
<i>R. officinalis</i>	Live	0	17.7 ± 7.7	24.1 ± 10.2	166.6 ± 95.2	↑
<i>U. parviflorus</i>	Live	0	0	0	182.1 ± 50.0	↑

For Peer Review

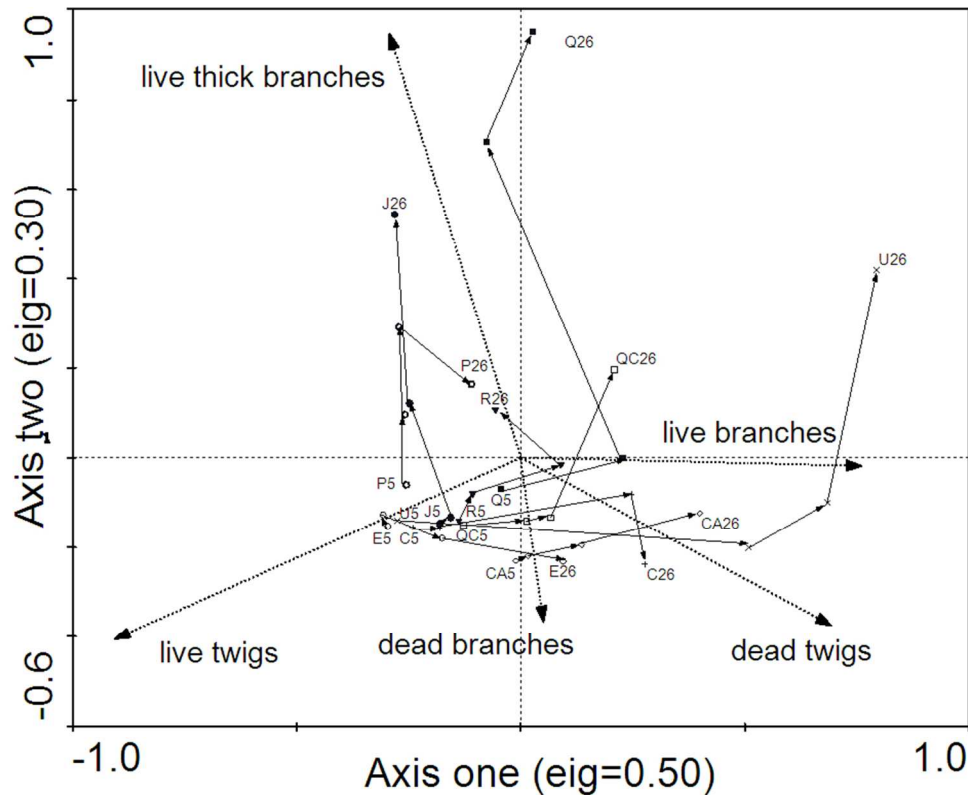


Fig. 1. PCA ordination biplot showing the ordination of species at different stand ages as compared to the first two PCA axes. The first two axes explain up to 80.6% of variance in species ordination. Arrows show the structural explanatory variables of live and dead fuel according to diameter class (twigs, branches and thick branches). PCA Axis 1 shows a marked negative correlation with live twigs ( $r=0.63$ ;  $P<0.0001$ ) and a marked positive correlation with dead twigs ( $r=0.33$ ;  $P=0.0003$ ). PCA Axis 2 correlates positively with thick branches ( $r=0.93$ ;  $P<0.0001$ ). For simplicity, numbers are used to indicate the first and last stand ages per species (5 and 26 years), but all the ages studied are shown, following the direction of the arrows (5, 9, 14 and 26 years). C= *C. clusii*, CA= *C. albidus*, E= *E. multiflora*, J= *J. oxycedrus*, P= *P. halepensis*, QC= *Q. coccifera*, Q= *Q. ilex*, R= *R. officinalis* and U= *U. parviflorus*.

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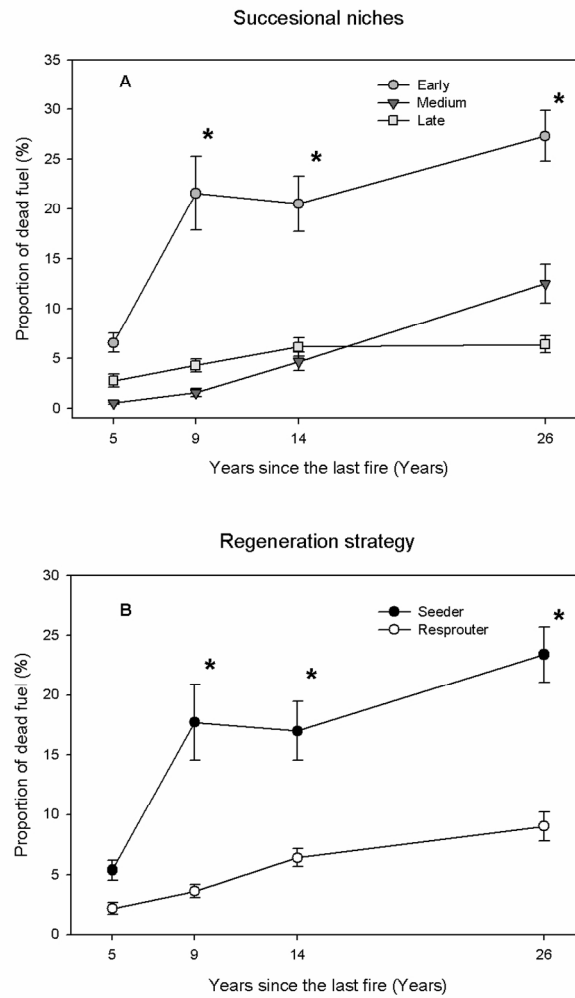


Fig. 2. Proportion of dead fuel for the different stand ages since the last fire between the functional groups: A) Successional niches (early-, medium- and late-successional species); B) Regeneration strategy (seeder and resprouter species). An asterisk denotes significant differences ( $p < 0.05$ ) for a specific age.  
209x296mm (150 x 150 DPI)

Ms. Ref. No.: PlaBio-2015-01-0025-RP

Title: Biological significance of the dead biomass retention trait in Mediterranean Basin species: an analysis between different successional niches and regeneration ability as functional groups.

Plant Biology.

Dear Prof. Heinz Rennenberg, Editor-in-Chief of Plant Biology:

Please find enclosed a copy of our revised manuscript in which all the suggestions made by Reviewers 1, 2 and 3 have been addressed. We list the changes that we have introduced into the ms. below

Reviewer #1: Specific comments.

-We have made all the minor suggestions, mainly about the English text and rephrasing sentences, raised by Reviewer 1. Nonetheless, we provide a point-by-point list of all the major changes made below:

- P2. “Regeneration ability” refers to the species capacity to regenerate. But the authors seem to be referring to the type of regeneration (sprouting or seeding).

**Response:** We agree with the Reviewer as this is unclear and confusing for the reader. In the title and throughout the ms., we have changed “Regeneration ability” to “Regeneration strategy”, which refers to the main (predominant) post-fire regeneration strategy (Resprouter/Seeder).

- P9, L3-4. Can you explain why is species a random factor? Why not site or species nested within site?

**Response:** See the response to Reviewer 2.

- P11, L4-6. You don't have results to support this sentence. Try to support it with the literature, if available.

**Response:** We have included a reference that supports this sentence (Moritz, M.A., Keeley J.E., Johnson E.A., Schaffner A.A. (2004). Testing a basic assumption of shrubland fire management: How important is fuel age? *Frontiers in Ecology and the Environment* **2**, 67-72).

-P11, L18. Note however that there's more to flammability than canopy structure. Species that do not retain dead foliage in the canopy can in turn accumulate flammable litter, and one of those species is *Quercus coccifera*.

**Response:** We have taken this reflection into account by including the note made by Reviewer 1 and a reference that supports this sentence: “species that do not retain dead foliage in the canopy can, in turn, accumulate flammable litter (Scarff & Westoby 2006)”. By also following the reviewer's suggestion, we have included a reference about Mediterranean ecosystems as an example (Curt T., Schaffhauser A., Borgniet L., Dumas C., Estève R., Gantaume A., Jappiot M., Martin W., N'Diaye A., Poilvet B. (2011) Litter flammability in oak woodlands and shrublands of southeastern France. *Forest Ecology and Management* **261**, 2214-2222.).

- P14, L5 Another thought for the discussion: dead fuel retention increases the likelihood of burning all year round, whereas low dead fuel limits the opportunities to burn.

**Response:** We have considered the suggestion made by Reviewer 1 about the effect of dead biomass retention on the seasonal probability of fire, and we have included a paragraph that explains this idea (see Page 12, Lines 15-20).

Reviewer #2:

We now explain in detail the comments raised by Reviewer 2 about our ms.

General comments.

- I suggest to re-focus the ms, emphasizing why stand flammability changes along the succession. This was not explicitly pointed in the hypothesis, but was evaluated in the analyses.

**Response:**

We partially agree with Reviewer 2 that the most relevant part of our study is the assessment of dead biomass produced in species with different successional niches. However, he/she suggests focusing our manuscript on how dead biomass changes during succession at the community level. We think that this suggestion goes beyond the scope of our paper. Variations at the community level during succession have been studied, as the reviewer pointed out, in Baeza et al (2011); however in this study, we focus our comparisons at the species level, and according to the successional niche that they occupy. This was our first hypothesis, which is clearly stated in the Introduction. This is the main part of our work and the Discussion, which focuses on species-specific responses in relation to different successional niches. The reviewer also suggests reducing the part about our second hypothesis (comparison between seeders and resprouters) because it is redundant in relation to the first hypothesis. However, we also think that our second hypothesis, proportion of dead biomass in relation to regeneration strategy, is interesting and novel to a certain extent. It is true, as suggested by the reviewers, that this hypothesis has been tested in previous studies (Cowand and Ackerly 2010, Saura-Mas et al. 2010). However, we tested the dynamics of dead biomass with time, and we observed some differences. Previous studies have focused only on a fixed point over time. These obtained results are novel and we consider them to be relevant enough for them to maintain in the manuscript because of the associated ecological and management implications. In any case, we have included a new paragraph in the Discussion that explains the possible interest of this analysis (page 11, Line 14-16).

-Another concern refers to the comparison of flammability between seeders and resprouters. In one hand, authors conduct this comparison because successional niches are strongly correlated with the post-fire regeneration strategy. This comparison is almost redundant with the comparison between successional stages, since all early species are seeders and all late species are resprouters. Secondly, this comparison was conducted to evaluate the hypothesis that traits increasing flammability should be more frequent in species with post-fire seeding.

**Response:** We partly agree with the reviewer since a set of traits shared between early and later successional species exists in nature; therefore the comparison made between successional niches and regeneration strategies might be redundant. Yet, as we previously explained, we believe that the comparison between regeneration strategies (seeder and resprouters) provides novel data given its temporal study. Thus leaving this



part could be of interest to readers. The reviewer also suggested that all the early species are seeders and that all the late species are resprouters. Yet in our study, species like *Erica multiflora* (resprouter) and *Rosmarinus officinalis* (seeder) are more typical of medium successional stages, as we described in the previously published conceptual vegetation dynamics model (Baeza et al. 2007). Anyway, in order to specify this possible drawback and the possible dependency of our results, we have included a new paragraph in the discussion where this relationship between the studied functional groups is clearly presented (Page 11, Line 7-10).

- Although resprouting and seeding abilities are negatively correlated in the Mediterranean Basin (Pausas & Verdú 2005), resprouting species might recruit after fire. In fact, *Erica multiflora* does (Paula et al. 2009). Therefore, this analysis was not the proper to test de second hypothesis.

**Response:** We do not agree with the reviewer because the main post-fire regeneration strategy of *Erica multiflora* is resprouting. In fact, in the plant trait database (Brot) (see Paula et al, 2009), 75% of the references made to *Erica multiflora* are classified as resprouter. By also following Pausas and Lavorel 2003, after fires, this species is classified as I+P-C-D+ parameters; that is: POSITIVE, individual persistence capacity (resprouting capacity); NEGATIVE, propagule persistence capacity (persisting in a seed form); NEGATIVE, competitive capacity (shade tolerance); POSITIVE, Dispersal capacity (short/long distance). In addition, the other three resprouter species (*Quercus ilex*, *Quercus coccifera* and *Juniperus oxicedrus*) are clearly considered non-seeding species after fire and resprouters.

Pausas and Lavorel, 2003. A hierarchical deductive approach for functional types in disturbed ecosystems. Journal of Vegetation Science 14: 409-416.

-Considering that the comparison of flammability between post-fire regeneration strategies is secondary in this study and it has been previously tested (Saura-Mas et al. 2010, Cowan & Ackerly 2010), I suggest to minimize the part of the ms designated to this issue.

**Response:** As explained above, we do not coincide with the reviewer's view. The studies referenced by the reviewer do not provide information about dead biomass retention with time in seeder/resprouter species, which is what we have shown in our ms., and which we believe is a relevant point.

Specific comments.

- P7 L18-19: Change by "the age of all the individuals per plot" by "the post-fire age of the plot". Consider this suggestion for the rest of the ms.

**Response:** We disagree with the reviewer about changing this sentence. We already knew plot age from checking the fire cartography. Specifically, we confirmed that the individuals selected for our study were of the same age as when the fire occurred. This is explained in more detail in the reference that we have included in the text (Baeza et al. 2011).

- P8 L9-19: How the PCA contribute to test the hypotheses of the ms?. If does not, then I recommend to remove it.

**Response:** As we explained in the Statistical analysis section, "Species with a similar biomass structure should be grouped near each other in the ordination space, which

should help identify any architectural pattern in biomass classes”. We consider that Figure graphically displays the temporal arrangement of the biomass structure per species in their different fractions (live twigs, dead branches, dead twigs, live branches). This may be helpful for showing a general idea of all the biomass components.

- P8 L19-24: Why authors do not compare also other physical descriptors of flammability (e.g., fine:coarse fuel ratio, dead fine fuel, etc...)?

**Response:** Our study looks at depth at the retention of dead biomass capacity; however, we believe that this suggestion has been covered in the PCA analysis, where all the biomass fractions are placed in time order per species.

- P9 L3-4: I suppose that the Species was included as random factor nested within Successional Niche or Regeneration Strategy, and the Individuals considered as pseudoreplicates. Currently, there are more sophisticated statistical tools to conduct LMM and the corresponding posthoc comparisons. Specifically, I recommend to use the packages lme4, lmerTest and lsmeans.

**Response:** Following the recommendations of Reviewer 2, we have modified our analysis. We firstly used the ANOVA function of the lmerTest package to test the global differences between the different successional niches and regeneration strategies after considering all ages together. This analysis also answers the concern about the data analysis raised by Reviewer 3. Secondly, the differences between successional niches and regeneration strategies for each specific age were analyzed by comparing the least-squares means with the “lsmeans” function. For this purpose, LMMs were fitted with age used as the discrete variable. These differences have also been shown in Figure 2A and 2B. Finally, as suggested by the reviewer, Species has been included as a random factor nested within Successional Niche or Regeneration Strategy. We think that these changes in the statistical analysis improve the paper and the interpretation is now much easier for readers.

- P9 L11-12: This result is not easily interpreted from Fig. 1, but from the Table S1 of the Supplementary Materials.

**Response:** We agree with the reviewer and have changed the sentence “At early ages (5 years), species are composed mainly of twigs (Fig. 1)” to “At early ages (5 years), species are composed mainly of live twigs (Fig. 1)”.

- P9 L22-P10 L10: Please rephrase, moving to the Discussion the sentences related to the interpretation of the results. For instance: “Dead biomass increase with post-fire age, being this increments stepper for early successional species, in such a way that differences between successional niches emerge in the older post-fire plot, where late species are those with lower dead biomass proportion”.

**Response:** We do not agree with the reviewer about moving the sentences about the interpretation of the results to the Discussion. In the Results, there is no sentence that matches the sentence the reviewer indicates. We believe that the Results section includes no sentence about the interpretation of the results.

- P10 L13-24: Please, rephrase emphasizing changes along the chronosequence, and the underlying mechanism.

**Response:** This is quite a vague suggestion and it is not clear what its contribution to improve the ms. actually is. Sentence P10 L13-24 corresponds to the first paragraph of

the Discussion where the main results of the study are found, and the details the reviewer refers to are considered throughout the Discussion.

- P11 L22 – P13 L4: This section is very speculative. The study conducted in the present ms does not contribute to know the selective pressure that shaped flammability traits. In addition, the information appears a little bit disordered, some sentences are contradictory to others (e.g., those about the relationship between standing dead biomass and heat released during fires) and the interpretation of some papers is not totally right (Pausas and collaborators in 2012 did not detect differences in dead biomass of *Ulex parviflorus* between sites differing in the fire regime, but in twig flammability).

**Response:** We do not coincide with the reviewer's opinion. In this section we discuss our results within the framed hypothesis, which suggests that "the evolution of post-fire regenerations traits coincides with the natural selection of enhancing flammability traits, such as retention of dead branches through increased post-fire recruitment". Our results provide clear evidence that early successional species retain more dead biomass than later species. Recent studies (Baeza and Roy 2008; Santana, Baeza and Blanes 2013) have indicated that different factors to heat exposure, such as daily temperature fluctuations, are involved in the seed germination of seeder species, such as *Ulex parviflorus*, and these mechanisms are often associated with early successional stages after fire or a different disturbance. We have also changed the sentence by including the reference to Pausas et al. (2012) in order to better interpret this reference (page 14, Line 3-4).

-P13 L13-17: Authors suggest that high dead biomass retention in early successional species would be a secondary consequence of the branching pattern of these species. I agree that intricate branching would decrease light interception efficiency, and thus diminish photo-oxidative damage by high light availability in recently burned communities. But if so, why plants would retain dead branches, considering their low influence in self-shading?

**Response:** In this section we have discussed different mechanisms that could be involved in dead biomass retention, among others the branching pattern of these species. Typical species of early successional stages share a set of traits, such as rapid growth, fast turnover of organs (leaves) and shade intolerance. When resources availability becomes scarce (light) in the lower strata, branches and leaves are no longer functional and accumulate as dead biomass, while later successional species are shade-tolerant and possess an effective self-pruning mechanism.

- P13 L22 - P14 L5: Species with low shoot:root ratio tend to be drought tolerant, whereas species with a big rooting system tend to be drought avoiders. In this sense, seeders shows more negative water potential at the turgor lost point (Vilagrosa et al. 2013), which do not support the hypothesis of a drought-driven origin of dead branch retention.

**Response:** We think that the reviewer has written this sentence wrongly (the species with a low shoot:root ratio and the species with a large rooting system are the same). We believe that the reviewer is mistaken when referring to "Species with a low shoot:root ratio tend to be drought-tolerant". The literature assumes that drought-tolerant species tend to have a high shoot:root ratio. If this is the case, then we agree with the reviewer that "Species with a high shoot:root ratio tend to be drought-tolerant, whereas species with a low shoot:root ratio tend to be drought avoiders". In our

ms, we did not compare the hypothesis that the reviewer puts forward: “a drought-driven origin of dead branch retention”. In our ms., the sentence that the reviewer refers to provides arguments that support our results, and this has been highlighted by Reviewer 3, who suggested new references to support our arguments (Saura-Mas S., Lloret F. 2013).

-P14 L6 – L16: The conclusion is speculative and not based in the results of the reviewed ms. To my understand, the results support the hypothesis that increasing flammability along the post-fire regeneration is explained by an increasing retention of dead biomass by early successional species. The eco-physiological underlying processes have not been evaluated in this study.

**Response:** We partly agree with the reviewer about the results obtained in our study: (i) seeders showed greater dead biomass accumulation than resprouters, and (ii) early-successional species accumulated more medium- and late-successional species. It was not our objective to evaluate eco-physiological processes; the eco-physiological underlying processes are commented on only in the Discussion to support our hypotheses.

Reviewer #3: Comments to the author.

-We have included the minor suggestions into the ms., along with the recommended references:

Bradshaw S. D., Dixon K.W., Hopper S.D., Lambers H., Turner S.R. (2011) Response to Keeley et al.: Fire as an evolutionary pressure shaping plant traits. *Trends in Plant Science* **16**, 405.

Saura-Mas S., Lloret F. (2007) Adult root structure of Mediterranean shrubs: relationship with post-fire regenerative syndrome. *Plant Biology* **16**, 147-154.

Saura-Mas S. (2008) Caracterització funcional d'espècies llenyoses mediterrànies amb diferents estratègies regeneratives post-incendi. Ph.D thesis. Universitat Autònoma de Barcelona.

Saura-Mas S., Lloret F. (2013) Leaf and shoot water content and leaf dry matter content of Mediterranean Woody species with different post-fire regenerative strategies. *Annals of Botany*, **99** (3), 545-554.

Material and methods

-Page 7 line 15: Please, specify why you sampled in winter and not in summer when there is the maximum dead biomass accumulation.

**Response:** We have included this sentence: “to avoid seasonal maximum dead biomass accumulation in summer”. Sampling in winter dead biomass is recommendable because production stabilizes during this period. Therefore, we can reduce the sampling error produced by the different rates of dead biomass production by species in summer.

- Page 7, lines 18-21: Please, specify why you calculated the age and how you extrapolated it for other species. Detail more this information, please.

**Response:** We have included the reference (Baeza et al. 2011), which provides details of the age of all the individuals by counting growth rings on the base of the stalk and by approximately coinciding with the years since the last fire for both the resprouter and seeder species.

-Results:

-Page 10, lines 6-8: Table 2 does not reflect that the analyses was considering all ages together, please, consider explaining with more details the difference between table 2 and table 1 analyses, maybe also in the data analyses section.

**Response:** See our response to Reviewer 2.

Table 1: Please, provide detailed information about which of the parameters are personal observations, and which of the parameters have been described in the literature. Specially, I consider it very important that successional niche information has been provided by some scientific published work

**Response:** We specified personal observation parameters (life span and time to maturity). The successional niche information was obtained from the conceptual model of the plant dynamics described by Baeza et al. 2007, as cited in Table 1.

-Figure 1. Please, describe here or in the results section, why there is only the representation of 5 and 26 years. Check this information in the legend, since all the 4 treatments appear in the text.

**Response:** We agree with Reviewer 3 that the interpretation of Figure 1 is somewhat complex. For this reason, we have added a sentence to the figure legend to explain that all the ages are shown, but only the initial age (5 years) and the final age (26 years) are denoted with a number for simplicity. However, we disagree with Reviewer 3 about changing the arrows to circles. We have attempt to what the Reviewer 3 suggests and the interpretation of the graph did not improve. We think that maintaining the arrows is the best version of the graph, where species are more clearly differentiated.

We would like to once again thank the reviewers for taking the time to review our manuscript, and for their many constructive and helpful comments. We also feel that the manuscript has substantially improved with the changes made, and we hope that this new version is now suitable for publication in *Plant Biology*.

Yours sincerely,

M. Jaime Baeza