Efecte de les pràctiques agricoles i del paisatge sobre la flora segetal dels secans mediterranis. Implicacions per a la conservació

Effect of farming practices and landscape on the segetal species of Mediterranean dry land arable fields. Implications for conservation

Roser Rotchés Ribalta

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Barcelona, maig de 2015

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Memòria presentada per la Roser Rotchés Ribalta per optar al grau de Doctora per la Universitat de Barcelona

Programa de Doctorat de Biodiversitat

Roser Rotchés Ribalta

Vist-i-plau dels directors de la tesi

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Barcelona, maig de 2015

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Roser Rotchés Ribalta
Sallent, 2015

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Efecte de les pràctiques agrícoles i del paisatge sobre la flora segetal dels secans mediterranis

Implicacions per a la conservació

Versió en català

Introducció general

1. Els sistemes agrícoles

1.1. Aspectes generals

Els ecosistemes agrícoles són sistemes antropogènics que depenen de l’activitat humana. El seu origen i el seu manteniment van associats a la intervenció de l’home, que ha transformat l’entorn per a la producció d’aliments, principalment. L’antiguitat i la naturalesa d’aquestes intervencions, ha donat lloc a un acoblament de les pràctiques agrícoles amb els sistemes que se’n generen. Per aquest motiu, l’agricultura no només té un paper clau en la provisió d’aliments per a les poblacions humanes, sinó també en el manteniment dels ecosistemes associats.

La terra agrícola representa una part molt important del paisatge, prop del 37,6 % de la superfície terrestre, el que situa l’agricultura com a principal ús del sòl (Food and Agriculture Organization of the United Nations 2013). Aquesta proporció és semblant en el context de Catalunya, on la superfície destinada a l’agricultura ocupa 1.125.268 ha, el 35,1 % del territori (Institut d’Estadística de Catalunya 2013). D’aquesta superfície, gairebé el 70 % és destinada a la producció de cultius herbacis extensius, dels quals més de dues terceres parts corresponen a conreus herbacis de secà (Institut d’Estadística de Catalunya 2013). Més del 30 % de la superfície agrícola de Catalunya (380.648 ha) es destina a la producció de cereal. Aquesta important extensió, tant a nivell mundial com regional, posa de manifest la rellevància d’aquests sistemes per a la conservació del territori.

1.2. La intensificació agrícola

La gestió agrícola ha anat canviant en relació amb els avenços tecnològics per atendre les demandes alimentàries d’una població mundial creixent. Aquests canvis van esdevenir més notables durant la segona meitat del segle XX, arran de l’anomenada ‘Revolució Verda’, amb la qual es va duplicar la producció mundial de cereal en tan sols 40 anys (Tilman et al. 2002); les dades disponibles per a l’estat espanyol així ho testimonien (Figura 1). No obstant, aquest augment de la producció ha anat en detriment de la qualitat ambiental en general (Tilman et al. 2001). Tant l'expansió de les àrees cultivades a costa d'ecosistemes naturals com la intensificació de les pràctiques agrícoles per aconseguir terres més productives (Foley et al. 2011) han estat clau per incrementar la producció, però també són la causa dels diferents impactes mediambientals. L'anàlisi dels canvis succeïts en l'agricultura arreu del món entre els anys 1985 i 2005 mostra que la producció ha augmentat un 28 % mentre que la superfície cultivada ho ha fet tan sols un 2,4 % (Foley et al. 2011). Per tant, la intensificació de la gestió agrícola, empesa per l'ús de varietats molt productives, la mecanització de les tasques agrícoles, l'aplicació de fertilitzants químics i l'ús de pesticides, entre d'altres, ha estat la principal responsable de l'increment de la producció agrícola (Matson et al. 1997), tal i com mostren les tendències a Espanya dels últims 50 anys (Figura 1).

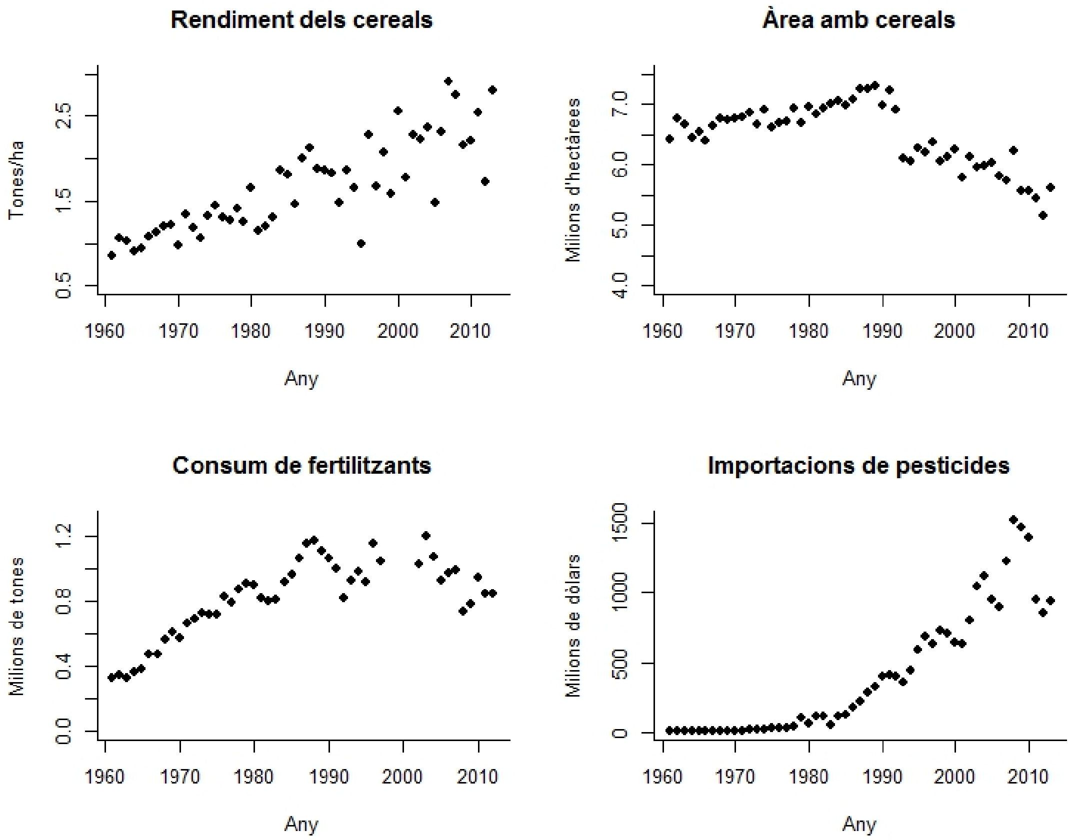

Figura 1: Tendències de l'agricultura a Espanya en els últims 50 anys. Rendiment dels cultius cerealistes, àrea sembrada amb cultius cerealistes, consum de fertilitzants i importacions totals de pesticides. Gràfics elaborats a partir de les dades de Faostat (2015).

La intensificació en l'ús de la terra agrícola que ha permès una concentració de la funció productiva, sovint, ha anat en detriment de la capacitat dels agrosistemes de subministrar béns i serveis. Les elevades aportacions de fertilitzants i pesticides tenen un fort impacte sobre la qualitat dels sòls i causen la contaminació de les aigües subterrànies (Tilman et al. 2002). La intensificació de les pràctiques agrícoles també exerceix importants canvis en la composició d'espècies de les comunitats biòtiques i és la causa de la remarcable disminució de la seva biodiversitat (Robinson & Sutherland 2002; Sans et al. 2013). Encara que els efectes de la intensificació agrícola s'han evidenciat arreu de manera general, els processos implicats operen a escales concretes tant a escala de paisatge, on la interacció amb l'entorn pot ser molt important (Robinson & Sutherland 2002; Tscharntke et al. 2005), com a escala de camp, on la gestió local hi té un paper primordial (Benton et al. 2003; Norton et al. 2009).

1.2.1. Intensificació agrícola a escala de paisatge

La intensificació que ha caracteritzat l'agricultura d'aquestes últimes dècades ha comportat canvis importants en els paisatges agrícoles. La concentració parcel·lària de la segona meitat del segle XX va anar associada a l'agregació de camps i a la reducció i simplificació estructural de molts dels hàbitats dels marges i altres formacions vegetals associades als conreus (Stoate et al. 2009). D'aquesta manera, el paisatge agrícola d'algunes zones ha patit un procés de simplificació: des de paisatges constituïts per un mosaic de conreus i marges i amb una elevada proporció d'hàbitats naturals i semi-naturals al seu voltant, a paisatges simples dominats per grans extensions cultivades i amb poca heterogeneïtat estructural (Robinson & Sutherland 2002; Roschewitz et al. 2005; Tscharntke et al. 2005; Baessler & Klotz 2006). La pèrdua dels hàbitats adjacents als conreus i la fragmentació dels hàbitats naturals per l'expansió de l'agricultura ha incrementat el risc d'extinció de les espècies amb poblacions més petites i aïllades, fet que ha comportat la pèrdua de biodiversitat (Roschewitz et al. 2005; Tscharntke et al. 2005).

1.2.2. Intensificació agrícola a escala de camp: gestió agrícola

La intensificació agrícola a escala de camp s'expressa principalment en la gestió dels conreus. La gestió intensiva que ha caracteritzat l'agricultura durant les últimes dècades en els països desenvolupats i en vies de desenvolupament conforma el que es coneix com agricultura convencional (Gliessman 2000). De manera general, la gestió convencional es caracteritza per elevades aportacions externes de fertilitzants orgànics i inorgànics i de pesticides químics, la mecanització que ha permès la llaurada intensiva, la simplificació de les rotacions, la sembra de grans extensions de monocultius i l’ús de llavors comercials lliures de males herbes, entre d’altres. Aquestes pràctiques no només afecten negativament la qualitat ambiental i la biodiversitat a escala de camp i de finca (Stoate et al. 2009) sinó que també tenen conseqüències sobre l’entorn de les explotacions agrícoles (Matson et al. 1997; Robinson & Sutherland 2002; Tilman et al. 2002; Foley et al. 2011).

Per tal de pal·liar aquests efectes de l’activitat agrícola, s’han desenvolupat i recuperat diferents models de gestió agrícola més respectuosos amb el medi ambient. Es tracta d’estratègies amb un enfocament més ecològic, que integren l’experiència de l’agricultura tradicional amb els coneixements científics i tecnològics actuals, amb l’objectiu de disminuir l’impacte mediambiental de les pràctiques convencionals (Matson et al. 1997). Pretenen preservar la fertilitat i la qualitat dels sòls i de l’entorn, a la vegada que potencien la biodiversitat i preserven l’equilibri ecològic natural (Food and Agriculture Organization of the United Nations 2002). Representen, en certa manera, un retorn a la gestió tradicional: basen la fertilització en l’aplicació de matèria orgànica i en la incorporació de lleguminoses i d’adobs verds en les rotacions de cultiu, i fan un control mecànic de les males herbes, entre d’altres. Malgrat es tracta d’una bona estratègia per a la preservació de la qualitat de l’hàbitat, l’agricultura ecològica no aconsegueix unes produccions tan elevades com l’agricultura convencional (Food and Agriculture Organization of the United Nations 2002; Seufert et al. 2012). És segurament per aquest motiu, entre d’altres (Musshoff & Hirschauer 2008) que la gestió ecològica és encara un model productiu minoritari, que tan sols representa el 2 % de la terra agrícola a Europa i no arriba al 10 % de la superfície agrícola de Catalunya. Tot i això, l’àrea ocupada per l’agricultura ecològica ha augmentat considerablement els darrers anys: entre el 2003 i el 2013 es va duplicar la superfície agrícola gestionada de manera ecològica a Catalunya (Consell Català de Producció Agrària Ecològica 2013).

1.2.3. Efectes de la intensificació agrícola sobre la biodiversitat

Un dels efectes més destacables de la intensificació agrícola tant a escala de parcel·la com de paisatge ha estat la pèrdua de biodiversitat (McLaughlin & Mineau 1995; Robinson & Sutherland 2002; Tilman et al. 2002). Els sistemes agrícoles, entesos com el mosaic d’àrees conreades i habitats naturals i semi-naturals, apleguen una proporció elevada de la biodiversitat en comparació amb altres sistemes terrestres (Pimentel et al. 1992). Però també són els sistemes on ha tingut lloc un declivi més acusat en els últims anys (Preston et al. 2003) (Figura 2). Convé, doncs, invertir esforços en la conservació d’aquelles espècies que habiten en àrees agrícoles a les quals la intensificació que ha tingut lloc les ha afectat més negativament (Pimentel et al. 1992). Es tracta d’un equilibri complex, que comporta la necessitat d’harmonitzar la producció agrícola i el manteniment integral de la diversitat dels diferents grups biològics que habiten els agrosistemes (Sans 2007; Zhang et al. 2007).

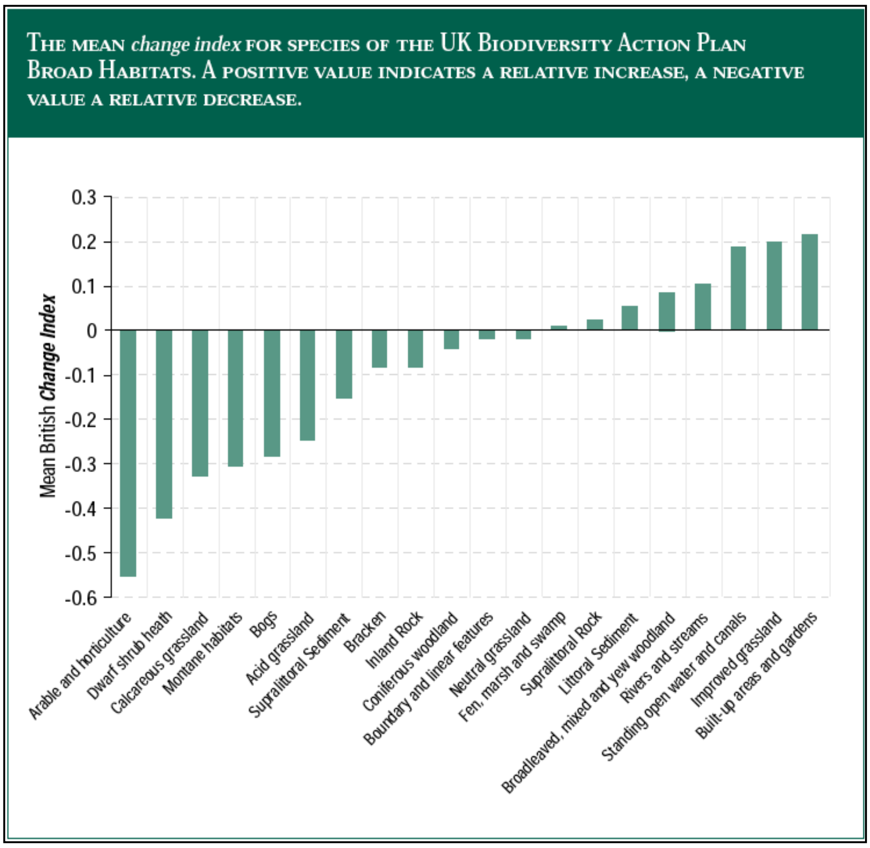

Figura 2: Índex del canvi mitjà de la diversitat d’espècies al Regne Unit segons l’hàbitat. Els valors en positiu indiquen un increment de la biodiversitat i, en canvi, els valors negatius indiquen pèrdua de la biodiversitat entre els mostrejos de 1930-1969 i 1987-1999 (Preston et al. 2002).

Tot i que en general es parla d’una pèrdua de biodiversitat, també han tingut lloc canvis importants en la composició específica de les comunitats. Mentre que hi ha una tendència per moltes espècies a disminuir la seva freqüència, d’altres troben unes condicions favorables i es fan dominants. A més, aquestes espècies que es fan més abundants són sovint espècies generalistes, el que condueix a una banalització de les comunitats biòtiques (Albrecht 2003). Per tant, la faceta més remarcable d’aquesta disminució de la biodiversitat als sistemes agrícoles ha estat la pèrdua de tàxons especialistes, que prosperen preferentment en sistemes agrícoles poc intensificats (Robinson & Sutherland 2002). Aquests efectes de la intensificació agrícola s’han descrit en diversos grups d’organismes, però sobretot en ocells, artròpodes i plantes que depenen d’alguna manera dels sistemes agrícoles (Robinson & Sutherland 2002).

2. La diversitat vegetal dels agroecosistemes

2.1. La diversitat vegetal associada als sistemes agrícoles

Les espècies arvenses són el conjunt de plantes que colonitzen els conreus tot i no haver estat sembrades de manera intencionada per l’agricultor. El desenvolupament i l’expansió de l’agricultura en els seus orígens va comportar la selecció d’una flora associada capaç de prosperar en aquests ambients. La diversitat vegetal d’aquests sistemes depèn, doncs, de l’establiment de les llavors que hi arriben per dispersió i de les que formen part del banc de llavors (Ryan et al. 2010), de manera que la diversitat d’un camp estarà determinada no només pels factors ambientals com el clima, el context paisatgístic i les característiques físiques del sòl, sinó també pel tipus de gestió agrícola (Roschewitz et al. 2005; José-María et al. 2010).

Les espècies arvenses, també anomenades ‘males herbes’, han estat considerades un factor limitant per a la producció del cultiu perquè hi competeixen directament pels mateixos recursos. S’ha calculat que poden causar pèrdues de fins al 30 % de la collita (Oerke 2006). A més, les plantes arvenses poden actuar com a hostes de plagues i malalties de les plantes cultivades i afectar negativament la producció i la qualitat de la collita (Masalles 2008). Per aquests motius, el control i la reducció de les poblacions de les males herbes ha estat un dels objectius principals de les estratègies de gestió agrícola. No obstant, el nombre d’espècies arvenses realment problemàtiques per a la producció del cultiu és limitat. La majoria de les espècies arvenses rarament provoca grans pèrdues de producció i, en canvi, contribueix de manera considerable a la biodiversitat d’aquests hàbitats (Albrecht 2003).

La biodiversitat és una part fonamental del patrimoni natural i està en relació amb la història dels usos del territori. Concretament, és difícil entendre els paisatges de la conca mediterrània sense l’agricultura. En conseqüència, la diversitat d’espècies arvenses té un valor intrínsec en formar part de la pròpia biodiversitat d’aquests sistemes, amb un important valor estètic i de conservació del paisatge agrícola, indicador d’una agricultura tradicional poc intensiva (Aboucaya et al. 2000; Clergue et al. 2005). A més, les espècies arvenses tenen un paper fonamental des d’una perspectiva funcional i ecològica. Les espècies arvenses proveeixen recursos alternatius per a pol·linitzadors, herbívors i granívors i són l’hàbitat i el refugi per a la fauna associada als cultius (Altieri 1999; Marshall et al. 2003; Clergue et al. 2005).

2.2. La flora segetal

Entenem per flora segetal el conjunt d’espècies arvenses que habiten preferentment als conreus cerealistes de secà. Diversos autors les han anomenat espècies arvenses característiques o obligades dels conreus de cereals (Sutcliffe & Kay 2000; Albrecht 2003; Romero et al. 2008b; José-María et al. 2010). La flora segetal inclou espècies que provenen del Pròxim Orient, des d’on van ser introduïdes per l’home amb l’expansió de l’agricultura, el que entenem per arqueòfits. Es tracta d’espècies no natives d’una regió geogràfica que van ser introduïdes i naturalitzades antigament, abans del 1500 dC (Pyšek et al. 2004). Ara bé, també inclou espècies que, malgrat ser autòctones, en l’actualitat es troben principalment en els camps de conreu.

Les espècies segetals, en general, són espècies anuals que requereixen les pertorbacions periòdiques característiques dels camps per poder re-establir-se anualment des del banc de llavors o des de propàguls arribats dels camps adjacents. La persistència d’aquestes plantes molt sovint depèn d’una fecunditat molt elevada i d’una capacitat de germinar que s’estén durant períodes relativament llargs (Masalles 2008). A la vegada, moltes d'aquestes espècies característiques dels camps cerealistes de secà són incapaces de persistir en ambients més competitius (Marshall 2009). La persistència de les seves poblacions a les nostres latituds, doncs, sembla lligada al manteniment de les explotacions agràries. Per aquestes raons aquestes espècies es consideren característiques de l'ordre fitosociològic *Secalietalia cerealis* Br.-Bl., 1936, que aplega les comunitats de males herbes dels camps de cereals d'hivern.

L'avaluació de la diversitat d'espècies vegetals en els camps de conreu típicament s'ha dut a terme considerant el conjunt d'espècies que no són sembrades intencionadament per l'agricultor (per exemple Roschewitz *et al.*, 2005; Hiltbrunner *et al.*, 2008). Ara bé, pot ser més adequat considerar tan sols les espècies que són característiques dels sistemes cerealistes de secà, ja que contràriament al nombre total d'espècies, aquest contingent no inclou aquelles espècies que apareixen freqüentment fora dels camps (Albrecht 2003). D'aquesta manera, espècies més generalistes com la calcida (*Cirsium arvense* (L.) Scop.) o l'apegalós (*Galium aparine* L.) no són considerades en l'avaluació de la diversitat.

2.2.1. Impacte de la intensificació agrícola en la flora segetal

Malgrat les espècies segetals estan adaptades a les pertorbacions periòdiques característiques de les pràctiques agrícoles, la intensificació de la gestió agrícola de les últimes dècades és la principal causa de l'important declivi de les seves poblacions (Sans et al. 2013). En general, les espècies característiques d'un hàbitat concret són més susceptibles de ser afectades pels canvis en les condicions ambientals i en l'ús de la terra que aquelles més generalistes (Albrecht 2003). En conseqüència, la intensificació agrícola ha suposat una disminució de les poblacions d'espècies segetals com la niella (*Agrostemma githago* L.), el blauet (*Centaurea cyanus* L.), la ballarida (*Hypecoum procumbens* L.), l'herba de l'amor (*Ranunculus arvensis* L.) o la rosella morada (*Roemeria hybrida* (L.) DC.) entre d'altres, a la vegada que altres espècies més generalistes han esdevingut més abundants (Masalles 2008; Romero et al. 2008b).

La pèrdua de diversitat d'espècies segetals ha estat descrita en diferents països europeus com Alemanya (Albrecht 1995), Dinamarca (Andreasen et al. 1996), Hongria (Kovács-Hostyánszki et al. 2011), el Regne Unit (Sutcliffe & Kay 2000; Robinson & Sutherland 2002) i Espanya (Cirujeda et al. 2011). A Catalunya aquesta qüestió també ha estat objecte d’estudi (Chamorro et al. 2007; Romero et al. 2008b), encara que no ha rebut l’atenció adequada (Sáez et al. 2011).

Són molts els canvis que han tingut lloc en la gestió dels conreus herbacis extensius en els darrers 60 anys, de manera que és difícil explicar adequadament els canvis que han ocorregut en la diversitat dels sistemes agrícoles simplement en relació amb un únic factor (Sutcliffe & Kay 2000). Malgrat això, s’atribueix una contribució significativa de la simplificació de les rotacions (Hyvönen & Salonen 2002) així com de les llaurades intensives (Mas & Verdú 2003; Santín-Montanyá et al. 2013) en la davallada de la diversitat vegetal als sistemes agrícoles. Per algunes espècies segetals en concret, però, la millora en la neteja de les llavors de la sembradura ha estat la causa de la disminució de les seves poblacions (Firbank 1988).

Tanmateix, es considera que l’ús d’herbicides per controlar les poblacions de males herbes ha estat una de les pràctiques agrícoles que més ha afectat les poblacions d’espècies arvenses (Robinson & Sutherland 2002; Potts et al. 2010). Des de la introducció de l’herbicida 2,4-D el 1946, el desenvolupament i l’ús de nous productes s’ha relacionat amb la davallada de la diversitat d’espècies arvenses (Heap 1997). A la vegada, l’elevada i constant aplicació d’herbicides als conreus ha donat lloc a l’aparició de fenotips resistents d’algunes espècies arvenses, moltes de les quals han esdevingut realment pernicioses (Heap 1997; Kudsk & Streibig 2003; Ulber et al. 2010).

L’increment en l’ús d’adobs, en particular dels de síntesi química, també pot haver exercit una pressió important sobre la diversitat d’espècies segetals. L’increment en la quantitat i disponibilitat de nutrients pot arribar a causar efectes directes de toxicitat sobre les plantes, però més generalment altera les interaccions competitives entre el cultiu i les espècies arvenses a favor del primer (Pyšek & Lepš 1991). En conseqüència, la flora segetal ha esdevingut un competidor pobre (Kovács-Hostyánszki et al. 2011). Aquest efecte s’ha intensificat amb el creixent ús de varietats de cultiu de creixement més vigorós. Els fertilitzants orgànics, per bé que poden suposar una entrada equivalent de nutrients al sistema, en general són menys concentrats i alliberen els nutrients més lentament, de manera que es redueixen els efectes negatius de la fertilització sobre la diversitat d’espècies arvenses (Robinson & Sutherland 2002).

Els canvis en l'estructura del paisatge agrícola també han contribuït en la pèrdua de diversitat d'espècies arvenses característiques dels conreus de cereals (Kovács-Hostyánszki et al. 2011; Solé-Senan et al. 2014). El procés de simplificació del paisatge ha afectat negativament les espècies segetals. Diverses investigacions han suggerit que amb aquest procés, d'una banda, s'ha reduït la proporció d'hàbitats semi-naturals i naturals que els poden servir de refugi i, d'altra banda, en homogeneïtzar el paisatge, s'ha facilitat la intensificació de la gestió agrícola.

2.2.2. Distribució de la flora segetal

Els efectes de la gestió agrícola i de l'estructura del paisatge sobre la flora segetal tenen un impacte diferencial al marge, a la vora i al centre del camp. L'impacte de la gestió agrícola és més remarcable al centre del camp i disminueix cap a la vora i els marges dels camps. D'altra banda, la importància de l'estructura del paisatge és més destacable als marges i a les vores dels camps i té menys efecte cap al centre dels camps (José-María et al. 2010; Kovács-Hostyánszki et al. 2011). Així doncs, la diversitat vegetal es concentra en els marges, i disminueix cap a l'interior dels camps. Ara bé, la flora arvense característica presenta un comportament diferent, ja que es concentra a les vores dels camps (Romero et al. 2008b; Fried et al. 2009; José-María et al. 2010) (Figura 3).

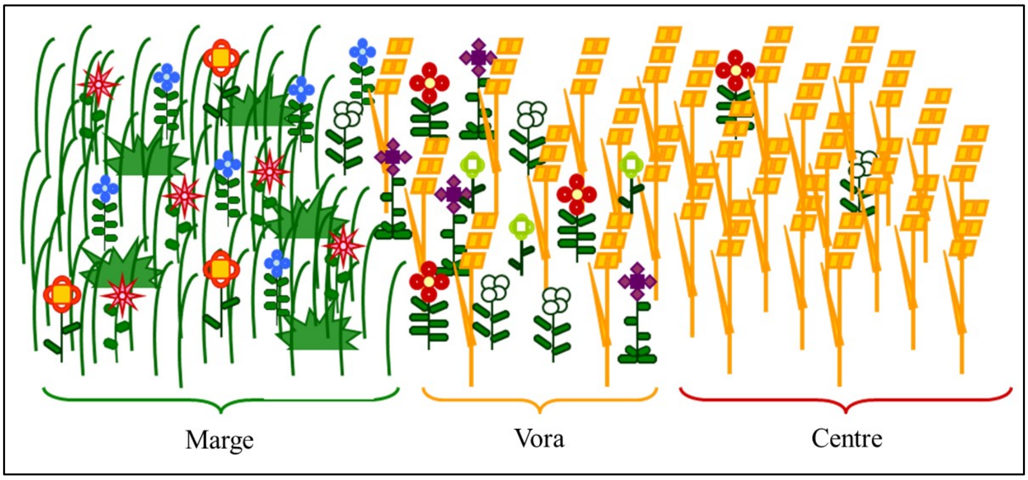

Figura 3: Esquema de la distribució de les espècies en els camps, des del marge fins al centre. La diversitat és més elevada als marges i més baixa cap al centre dels camps. Tot i això, a la vora dels camps hi habita de manera preferent la flora segetal o característica dels hàbitats cerealistes de secà que requereix les pertorbacions associades a la gestió del cultiu de cereals a la vegada que es beneficia de la menor eficàcia de les pràctiques agrícoles com la fertilització i el control de la flora arvense en aquesta àrea del camp.

Les vores dels camps són les franges de terra cultivada més externes del camp, en contacte amb els marges formats per hàbitats naturals o semi-naturals (Marshall & Moonen 2002). A les vores dels camps les pràctiques agrícoles són menys eficaces a causa de la dificultat de la maquinària per accedir-hi. Per aquest motiu, la quantitat rebuda de fertilitzants i herbicides és generalment menor, la llaurada no és tan regular i hi ha més errors de sembra, fets que redueixen la pressió competitiva del cultiu sobre les espècies arvenses (Kleijn & van der Voort 1997; Dutoit et al. 2007; Romero et al. 2008b).

2.2.3. Les espècies segetals rares

L'impacte de la intensificació agrícola d'aquestes últimes dècades s'ha fet molt evident en la important davallada de les poblacions d'espècies segetals fins al punt de convertir-ne algunes en molt poc freqüents o fins i tot extingides regionalment (Baessler & Klotz 2006; Fried et al. 2009; Cirujeda et al. 2011; Storkey et al. 2012). Molts països europeus ja han tingut en compte aquestes espècies arvenses menys freqüents en les respectives llistes vermelles (Schnittler & Ludwig 1996; Moser et al. 2002; Cheffings & Farrel 2005; Colling 2005; Türe & Böcük 2008; Moncorps & Siblet 2012). Aquesta consideració els atorga un cert valor, que constitueix la base per a la majoria d'estratègies de conservació. En alguns països, les espècies segetals rares han estat considerades espècies prioritàries per a la conservació en certs plans d'acció, com el del Regne Unit (Preston et al. 2003) o el de França (Aboucaya et al. 2000).

Malauradament, a la regió mediterrània la raresa d'aquestes espècies ha atret comparativament poc l'atenció dels investigadors i conservacionistes. Per exemple, les espècies arvenses no són considerades al *Llibre vermell de les plantes vasculars endèmiques i amenaçades de Catalunya* (Sáez et al. 2011) pel fet de dependre del manteniment dels sistemes antròpics i, algunes d'elles, ser introduïdes. L'única valoració de la seva freqüència en el territori ha estat duta a terme en les flores locals, amb criteris poc sistemàtics i considerant la flora regional en conjunt. En cap cas hi ha hagut al darrere una anàlisi detallada de l'estat de les seves poblacions. Malgrat l'evident pèrdua d'espècies segetals a Espanya i a Catalunya (Chamorro et al. 2007; Cirujeda et al. 2011) no s'han proposat estratègies per a la conservació d'espècies segetals, o tan sols se n'han proposat en un àmbit local (Mateu & Segarra 1998).

En el context europeu s’han implementat el que s’anomenen esquemes agro-ambientals (‘agri-environmental schemes’ o AES) (European Environment Agency 2004), els quals obliguen a tots els estats membres a desenvolupar i implementar programes agro-ambientals basats en incentius monetaris amb l’objectiu de mitigar l’impacte negatiu de la intensificació agrícola. Aquests programes no només estan destinats a la preservació de la biodiversitat sinó també al manteniment i a la millora de les condicions mediambientals. L’agricultura ecològica ha estat promoguda com una mesura agro-ambiental en alguns països (Kleijn et al. 2006) perquè aquests sistemes de gestió menys intensius poden afavorir la persistència de les espècies arvenses característiques dels cultius cerealistes (Kleijn et al. 2006; Rundlöf et al. 2009; Gabriel et al. 2010; Kolářová et al. 2013). Tot i això, l’efectivitat de les mesures agro-ambientals en relació amb la conservació de la diversitat vegetal a l’àrea mediterrània s’ha vist limitada pels efectes compartits de la complexitat del paisatge i la intensitat de gestió a diferents escales (Concepción et al. 2008).

La conservació de les espècies arvenses és objecte de controvèrsia, ja que el seu hàbitat òptim, els camps de cereals, és subjecte a uns interessos productius i econòmics, el que condueix al conflicte permanent entre la producció agrícola i la conservació d’espècies (Fried et al. 2009; Foley et al. 2011). Tot i això, és necessari destinar esforços a la conservació de la diversitat biològica en els sistemes agrícoles. Per aquest motiu convé avaluar l’estat de les poblacions d’espècies segetals que són o han esdevingut rares a l’àrea mediterrània, i conèixer millor la seva resposta davant les pràctiques agrícoles per poder establir unes mesures de conservació adequades.

3. Disseny de l’estudi

Aquesta tesi pretén millorar el coneixement existent sobre les espècies arvenses característiques dels camps de cereals de secà (comunitats de l’ordre fitosociològic *Secalietalia cerealis* Br.-Bl., 1936) i avaluar els motius pels quals algunes d’aquestes espècies han esdevingut poc freqüents en aquests hàbitats. Amb aquesta finalitat, l’estudi s’ha estructurat al voltant de dos eixos principals, que pretenen: 1) analitzar la freqüència de les espècies arvenses característiques i especialment d’aquelles que han esdevingut rares en relació a la gestió dels cultius cerealistes de Catalunya (*Capítols 1* i *2*) i 2) avaluar els efectes que han pogut tenir diferents pràctiques agrícoles en l'eficàcia biològica d'algunes d'aquestes espècies (*Capítols 3* i *4*).

3.1. Anàlisi de la freqüència de les espècies arvenses característiques i rares als conreus cerealistes de secà de Catalunya

3.1.1. Àrea d'estudi: les àrees cerealistes de secà de Catalunya

L'estudi de l'estat de les espècies segetals es va dur a terme el 2011 a la zona cerealista de Catalunya, majoritàriament a la Depressió Central. L'àrea d'estudi ocupa aproximadament 100 km × 80 km (de 41º 22' a 42º 06' N i de 0º 59' a 2º 12' E), i té una altitud mitjana (± SE) de 558 ± 30 msnm (Figura 4). El clima de la zona és mediterrani continental, amb temperatures mitjanes anuals (± SE) de 12.6 ± 0.2 ºC (entre 9.5 ºC i 14.9 ºC) i precipitacions mitjanes anuals de 637 ± 21 mm (de 416 a 868 mm) (Ninyerola et al. 2005). Aquesta variabilitat respon al fet que l'àrea estudiada comprèn un gradient climàtic, amb unes condicions més seques cap al sector més occidental. De manera general, l'àrea estudiada es troba sobre margues i gresos calcaris, substrats sobre els quals es desenvolupen sòls de caràcter bàsic.

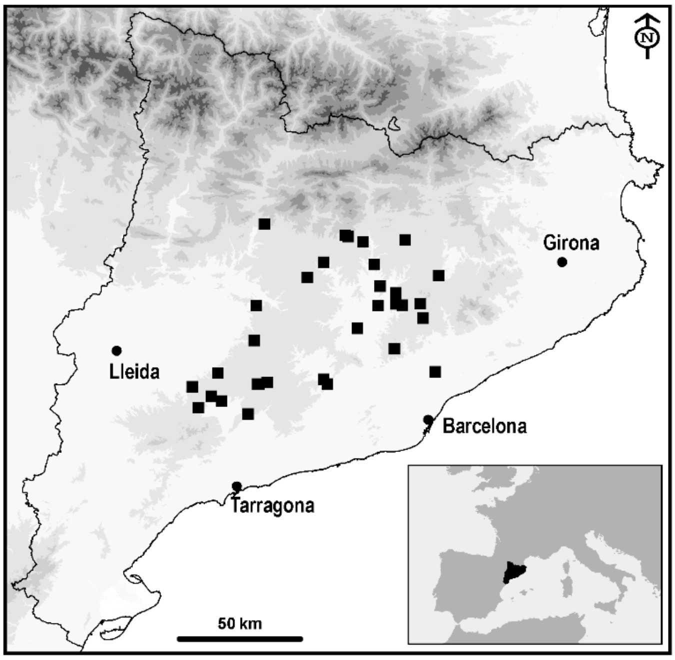

Figura 4: Localització de l'àrea d'estudi a Catalunya. Els quadrats indiquen les localitats mostrejades. Els tons en gris indiquen l'elevació cada 500 m.

El paisatge agrícola d'aquesta zona és un mosaic de conreus amb fragments d'hàbitats semi-naturals i naturals i zones urbanes, industrials i viàries. Els hàbitats naturals de l'àrea estudiada inclouen pinedes (*Pinus halepensis* Mill. i *P. nigra* Arnold subsp. *salzmannii* (Dunal) Franco), alzinars (*Quercus ilex* L.) i rouredes (principalment *Q. faginea* Lam.), matollars, pastures perennes i vegetació de ribera de cursos d'aigua intermitents. Als marges dels camps també hi són freqüents bardisses, llistonars i fenassars i diverses comunitats ruderals. L'agricultura representa un dels usos del sòl majoritaris en aquesta àrea, dominada per cultius cerealistes de secà principalment d'ordi (*Hordeum vulgare* L.) i blat (*Triticum aestivum* L.) i en menor grau de civada (*Avena sativa* L.) i espelta (*Triticum aestivum* L. subsp. *Spelta* (L.) Thell.). Aquests cultius conformen el mosaic agrícola juntament amb cultius farratgers, especialment d'alfals (*Medicago sativa* L.) i altres cultius anuals de secà com la colza (*Brassica rapa* L. subsp. *oleifera* (DC.) Metzg.) i el gira-sol (*Helianthus annus* L.), però també amb vinyes (*Vitis vinifera* L.) i camps d'ametllers (*Prunus dulcis* L.) i d'oliveres (*Olea europaea* L. var. *europaea*), entre d'altres (Departament d'Agricultura Alimentació i Acció Rural. Generalitat de Catalunya 2009).

Les comunitats d'espècies arvenses dels cultius anuals s'han considerat tradicionalment dins l'ordre fitosociològic *Secalietalia cerealis* Br.-Bl., 1936. A l'àrea d'estudi, però, tan sols hi trobem aquelles comunitats de caràcter més basòfil. Es tracta de comunitats dominades per espècies anuals com la rosella (*Papaver rhoeas* L.), el mill de sol petit (*Lithospermum arvense* L. subsp. *arvense* = *Buglossoides arvensis* (L.) I. M. Johnst. subsp. *arvensis*), les agulles de pastor (*Scandix pecten-veneris* L. subsp. *pecten-veneris*), l'herba de l'escorpí (*Coronilla scorpioides* (L.) Koch), l'unflabou (*Bupleurum rotundifolium* L.) o el cospí (*Caucalis platycarpos* L.), entre d'altres.

3.1.2. Disseny experimental. Capítols 1 i 2

Es va seleccionar un total de 304 camps de 32 localitats (Figura 4). Només es van seleccionar finques de gestió ecològica (una finca per localitat) perquè d'aquesta manera s'incrementava la probabilitat de trobar aquestes espècies segetals (Romero et al. 2008b; José-María et al. 2010). Fora de la Depressió Central Catalana es van considerar algunes finques amb conreus cerealistes gestionats de manera ecològica. Les localitats recullen un gradient de complexitat del paisatge, estimat a partir del valor mitjà del percentatge de terra cultivable (PAL) en àrees circulars d'1 km de radi al voltant de cada camp per cada localitat. L'estructura del paisatge per aquestes localitats varia des de paisatges estructuralment simples, amb valors alts de PAL (al voltant del 97 % de terra cultivable) a paisatges més complexos, amb predominança d'hàbitats naturals i semi-naturals (tan sols un 8 % de terra cultivable) i, per tant, on la pressió exercida per l'agricultura és més baixa (Figura 5).

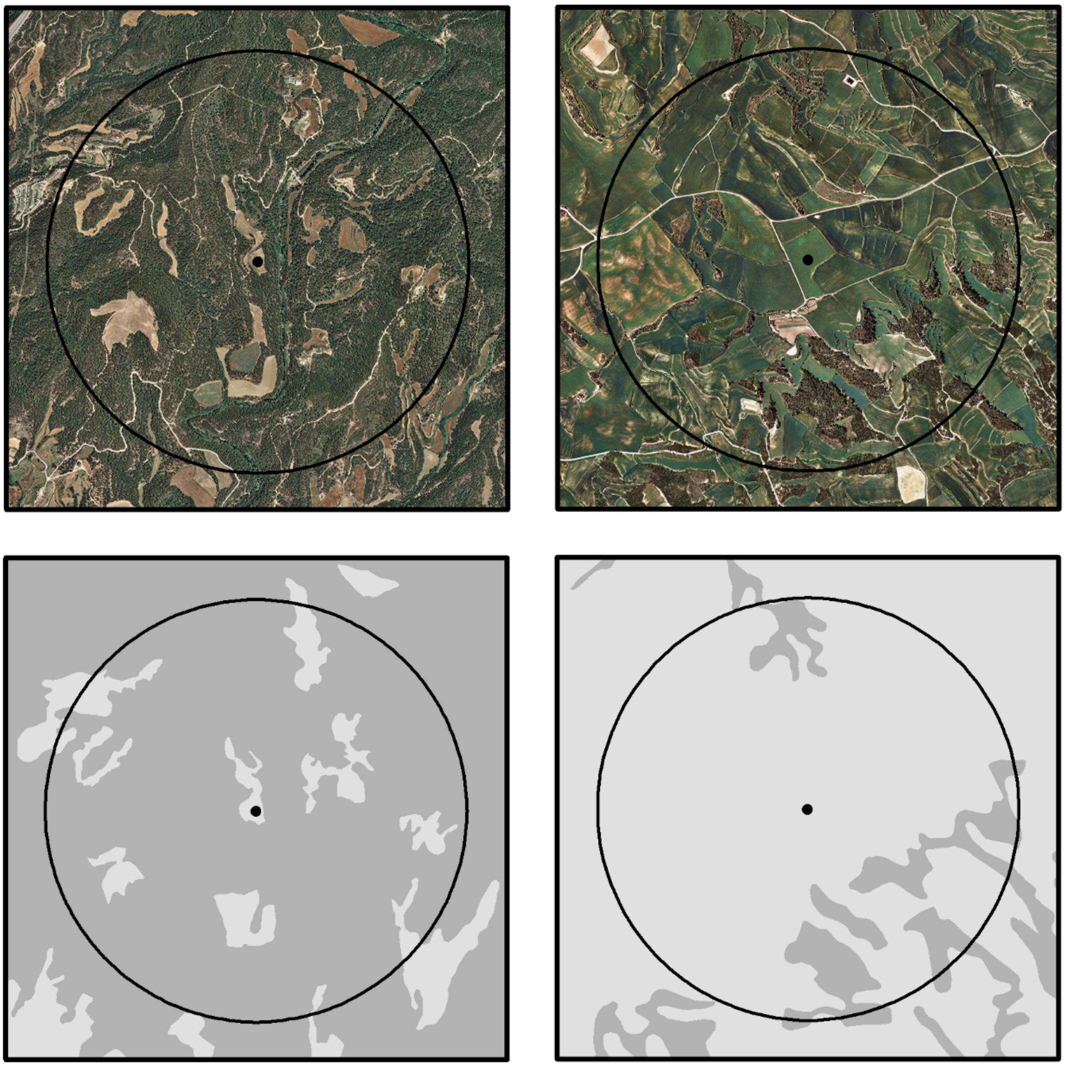

Figura 5: Exemples de camps mostrejats (indicat amb un punt negre) representatius d'un paisatge complex (esquerra) i simple (dreta). A la part superior es mostren els ortofotomapes de les àrees d'1 km de radi al voltant de cadascun d'aquests camps. En les imatges inferiors es mostren els mateixos camps amb la Cartografia dels Hàbitats de Catalunya convertida a un mapa binari, amb els hàbitats agrícoles en gris clar i els hàbitats naturals i semi-naturals en gris fosc.

La proporció de terra ocupada per conreus herbacis extensius (‘Percentage of arable land’, PAL) és una de les mesures més utilitzades com a indicador de la intensificació agrícola a escala de paisatge. Aquesta variable correlaciona amb altres descriptors del paisatge com la diversitat i la fragmentació dels hàbitats, la densitat dels marges o l’extensió d’hàbitats semi-naturals (Roschewitz et al. 2005). A més, aquestes correlacions es troben en paisatges ben contrastats com són els del nord i el centre d’Europa i els mediterranis (Roschewitz et al. 2005; Gabriel et al. 2005; Romero et al. 2008a).

A cada finca es van seleccionar els camps que havien estat sembrats durant la temporada immediatament anterior al mostreig amb els cultius anuals que formen part de la rotació típica dels conreus herbacis extensius de secà: cereals per a gra i per a farratge, alternats amb les lleguminoses també per a gra o per a farratge i els policultius que contenen barreges de cereals i lleguminoses. Els conreus plurianuals es van evitar a causa de la seva heterogeneïtat en el moment de sembra. Es van mostrejar entre 4 i 12 camps per localitat en funció del nombre de camps per finca que reunien les condicions dels criteris de selecció. Per tal de maximitzar la detecció de les espècies segetals rares, el mostreig es va restringir a les vores dels camps. Se’n va avaluar la presència de la flora segetal en punts de mostreig separats entre ells aproximadament 80 m al llarg de la vora del camp (Figura 6). Per tant, depenent de la mida del camp, es van mostrejar un mínim de tres i un màxim de deu punts per camp. A cada un dels punts de mostreig es van inventariar les espècies segetals que hi apareixien, en una àrea de 2 m × 5 m paral·lela al marge. També es van anotar una sèrie de característiques del punt de mostreig com el recobriment del cultiu i de les espècies arvenses totals, així com el tipus d’hàbitat del marge adjacent agrupats en les categories: vegetació llenyosa, prats i pastures, vegetació ruderal i altres.

La informació de la gestió de cada parcel·la es va obtenir mitjançant entrevistes que es van realitzar personalment als agricultors. Amb aquestes entrevistes es va adquirir la informació sobre la gestió de l’última temporada, en concret el tipus de llaurada, el cultiu sembrat, l’origen de la llavor, el moment i la densitat de sembra, el tipus i la quantitat de fertilitzant, el tipus de control de les males herbes i si s’hi va fer pasturar el bestiar. També es va recollir informació dels anys des de la conversió a agricultura ecològica i dels cultius

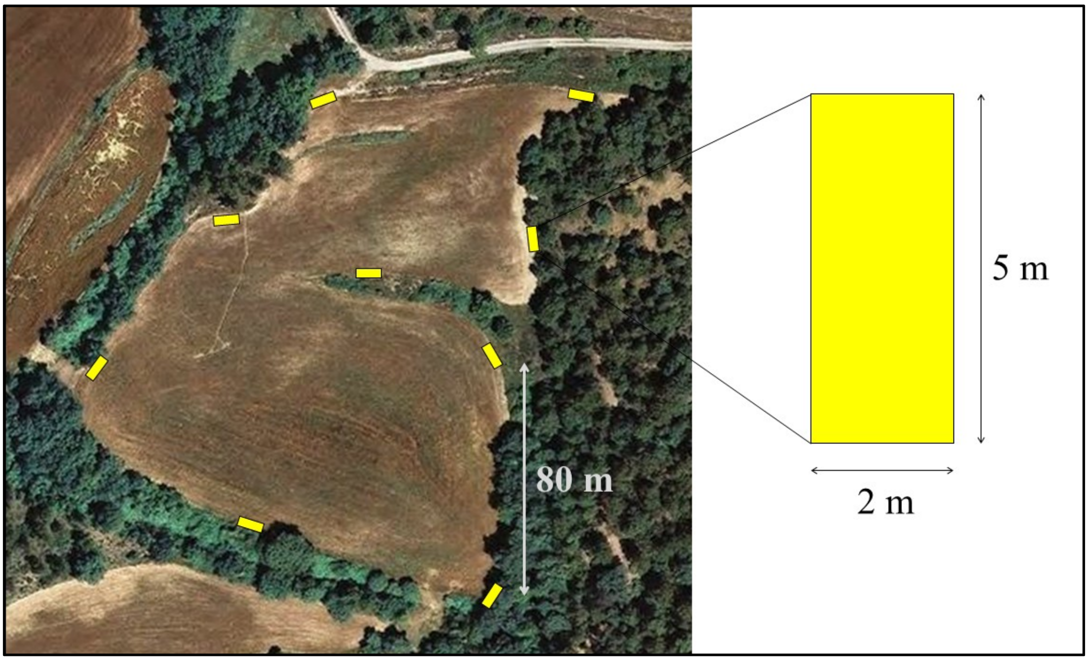

Figura 6: Esquema de la distribució dels punts de mostreig en un camp. Els punts de mostreig es van situar a la vora dels camps, separats entre ells 80 m. A cada punt de mostreig es va avaluar la presència de les espècies segetals en àrees de 2 m × 5 m.

sembrats durant els darrers cinc anys. Es va caracteritzar la gestió a nivell de finca en termes de l'orientació productiva de la finca (especialista en producció vegetal o amb cultius i ramaderia) i la proporció de camps amb cereals en l'any en curs.

L'estructura del paisatge es va caracteritzar mitjançant la proporció del paisatge circumdant ocupada per conreus herbacis extensius (PAL) en àrees circulars d'1 km de radi al voltant de cada camp. També es va definir l'estructura física dels camps, amb mesures de l'àrea i del perímetre dels camps. Aquestes mesures de l'estructura dels camps i del paisatge es van obtenir a partir d'ortofotomapes en color i de la Cartografia dels Hàbitats de Catalunya (Carreras & Diego 2004) mitjançant el programari d'ArcGIS (versió 9.2). L'estructura del paisatge de les finques també es va tenir en compte, a través del PAL mitjà al voltant dels camps per finca i la distància mitjana entre els camps de la finca.

Les dades obtingudes en els inventaris d'aquest mostreig van permetre avaluar l'efecte de les diferents pràctiques agrícoles ecològiques a nivell de camp i de finca (localitat) sobre la diversitat de les espècies arvenses segetals i d'aquelles que es consideren rares, considerant també les correlacions amb la complexitat del paisatge i amb l'àrea i la forma dels camps (*Capítol 1*). Aquesta diversitat es va analitzar, en el *Capítol 1*, mitjançant la descomposició multiplicativa de la diversitat (Allan 1975), que permet fer una partició de la diversitat total en components independents (Jost 2007). D'aquesta manera la riquesa específica d'una finca (diversitat γ) es descompon en la contribució mitjana de cada camp (diversitat α) i en la diversitat deguda a la divergència en la composició d'espècies entre els camps d'una localitat (diversitat β, calculada com a $\beta = \gamma / \alpha$). Paral·lelament, es va dur a terme una anàlisi més detallada de l'efecte de la gestió agrícola i de l'estructura dels camps i del paisatge, sobre la probabilitat de trobar les espècies segetals rares en un camp, així com les característiques del punt de mostreig i de l'hàbitat adjacent que afavoreixen la presència d'aquestes espècies (*Capítol 2*). Aquestes anàlisis també es van dur a terme per cadascuna de les espècies segetals rares més freqüents, per tal de conèixer com aquestes espècies responen individualment davant les variables considerades i si existeixen uns patrons comuns de resposta (*Capítol 2*).

3.2. Avaluació dels efectes de certes pràctiques agrícoles sobre l'eficàcia biològica d'algunes espècies segetals

3.2.1. Disseny experimental. Capítols 3 i 4

S'han portat a terme dos estudis sota condicions controlades per a avaluar els efectes de determinades pràctiques agrícoles sobre el creixement i la capacitat reproductiva de les espècies segetals. En aquests estudis s'ha avaluat l'impacte de l'aplicació d'herbicides (*Capítol 3*) i de la fertilització (*Capítol 4*) sobre diverses parelles d'espècies segetals, cada parella d'una família diferent. Es van seleccionar les espècies de cada família de manera que diferissin en la seva freqüència en el territori estudiat, essent una més rara i una més comuna per cada família. Aquest tipus d'assaig comparatiu permet tenir en compte el fet que plantes filogenèticament properes poden respondre de manera semblant a les condicions ambientals, el que fa més fiable la comparació entre espècies segetals comunes i rares.

En el *Capítol 3*, es va analitzar l'efecte dels herbicides 2,4-D i tribenuron aplicats a sis dosis diferents a part dels controls sobre quatre parelles d'espècies segetals de famílies diferents, incloent una espècie rara i una més comuna de cada família (Apiàcies: *Bupleurum rotundifolium* L. i *Scandix pecten veneris* L. subsp. *pecten-veneris*; Brassicàcies: *Neslia paniculata* (L.) Desv. subsp. *thracica* (Velen.) Bornm i *Rapistrum rugosum* (L.) All.; Papaveràcies: *Papaver argemone* L. i *P. rhoeas* L.; Rubiàcies: *Asperula arvensis* L. i *Galium aparine* L. subsp. *spurium* (L.) Simonk). En aquest experiment es va avaluar el grau d'afectació del creixement i de la capacitat de produir llavors de les espècies segetals testades a curt termini (28 dies després de l'aplicació dels herbicides) i a llarg termini (48 o 55 dies després de l'aplicació dels herbicides), així com determinar la capacitat de recuperació d'aquestes espècies després de l'aplicació dels herbicides a dosis subletals. També es va analitzar si les diferències entre espècies tenen una correlació amb les seves freqüències al territori.

Atès que la fertilització pot tenir efectes molt importants sobre el creixement i la reproducció de les espècies segetals mitjançant l'alteració de la competència entre l'espècie cultivada i les espècies arvenses, l'assaig per caracteritzar l'efecte de la fertilització es va dur a terme juntament amb l'avaluació de l'efecte de la competència del cereal. En el *Capítol 4* s'avalua de manera conjunta l'efecte de l'aplicació de diferents tipus de fertilitzants (orgànic i mineral) a diferents dosis (alta i baixa) sobre quatre parelles d'espècies segetals comunes i rares, també aparellades segons les famílies a les que pertanyen (les mateixes parelles que al *Capítol 3* excepte les Apiàcies, de les quals es va substituir *Bupleurum rotundifolium* L. per *Bifora testiculata* (L.) Spreng.), creixent amb competència o no de cereal (Figura 7).

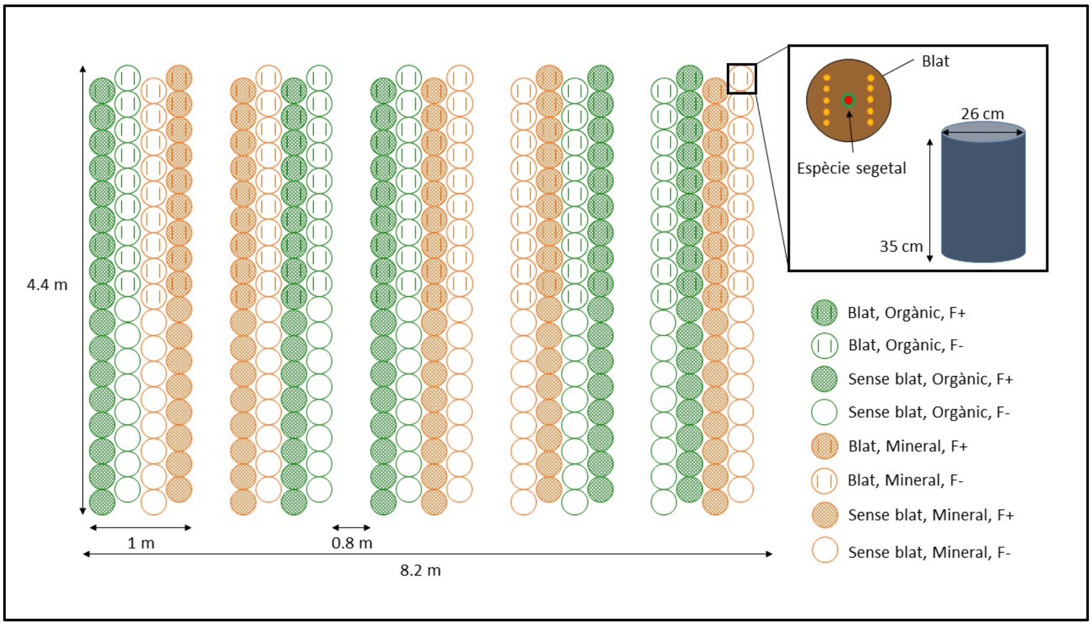

Figura 7: Esquema del disseny experimental emprat per a avaluar els efectes del tipus (orgànica i mineral) i de la dosi (alta, F+ i baixa, F-) de fertilitzants aplicats i de la competència (amb blat i sense blat) amb el cultiu (*Capítol 4*), on es representa la distribució dels testos segons el tractament. A dalt a la dreta, es mostra un test en vista superior i lateral, amb la mida dels testos i la disposició de les plantes de blat i les espècies segetals al substrat.

Objectius

1. Objectius generals

L'objectiu principal d'aquesta tesi doctoral és estudiar l'estat de les poblacions de les espècies arvenses característiques dels secans mediterranis en relació amb les pràctiques agrícoles i amb l'estructura del paisatge, i avaluar els motius pels quals algunes d'elles poden haver esdevingut rares en aquests hàbitats agrícoles. Amb aquesta finalitat, l'estudi ha estat dividit al voltant de dues línies d'investigació principals que són complementàries, una de caràcter observacional i l'altra experimental, amb la intenció de:

- Determinar els efectes de les diferents pràctiques agrícoles i la contribució de l'estructura del paisatge sobre la situació de les espècies arvenses característiques i especialment d'aquelles considerades rares en els cultius de cereals de Catalunya. Per això s'ha avaluat la riquesa i la presència d'aquestes espècies en les vores dels camps de gestió ecològica de la regió cerealista de Catalunya.

- Avaluar la resposta d'algunes espècies arvenses característiques dels conreus cerealistes de secà davant certes pràctiques agrícoles, en concret l'aplicació d'herbicida i la fertilització juntament amb la competència amb el cereal, i comprovar fins a quin punt el grau de raresa pot dependre de les diferències en l'eficàcia biològica d'aquestes espècies en resposta a aquestes pràctiques agrícoles.

2. Objectius específics

Els objectius específics de cadascun dels capítols en que s'ha estructurat aquesta tesi doctoral es detallen a continuació:

Capítol 1:

i) Estudiar els efectes de les pràctiques agrícoles actuals i passades (en els darrers 5 anys), de la mida i la forma dels camps i de l'estructura del paisatge sobre la riquesa d'espècies segetals i d'aquelles espècies considerades rares (diversitat α), en camps de gestió ecològica.

ii) Determinar els efectes de la gestió a nivell de localitat, entesa com el conjunt de pràctiques que afecten a la finca en la seva totalitat, a part dels efectes de les variables descriptives del paisatge circumdant, sobre la diversitat de la localitat (diversitats γ i β) d’aquests conjunts d’espècies.

Capítol 2:

i) Avaluar els efectes de les pràctiques agrícoles, de l’estructura dels camps i del paisatge i de les condicions locals sobre la presència d’espècies segetals rares a les vores dels camps ecològics de la Depressió Central Catalana.

ii) Determinar els patrons comuns en les respostes d’espècies segetals rares particulars a les diferents variables de gestió i de l’entorn testades.

Capítol 3:

i) Examinar els patrons de sensibilitat d’algunes espècies segetals a dos dels herbicides més utilitzats en els conreus de cereals de la regió mediterrània (el tribenuron i el 2,4-D) segons la biomassa aèria total, la biomassa reproductiva i el nombre de llavors a curt i a llarg termini.

ii) Determinar el grau i el temps de recuperació, en termes de llargada de la tija i del nombre de llavors produïdes, de les plantes afectades per dosis creixents d’aquests herbicides.

iii) Avaluar si la sensibilitat a l’aplicació d’aquests herbicides és més alta per espècies segetals rares que per les espècies més comunes relacionades filogenèticament.

Capítol 4:

i) Analitzar els efectes de la fertilització, en funció del tipus i de la dosi de fertilitzant, sobre el creixement d’algunes espècies arvenses característiques dels camps de cereals i com aquests efectes es veuen afectats per la competència amb el cereal.

ii) Determinar si el moment de floració de les espècies segetals és influït per les diferents dosis i tipus de fertilitzants interactuant amb la competència amb el cereal.

iii) Comparar l’habilitat competitiva de les espècies segetals rares amb les espècies més comunes emparentades filogenèticament sota els diferents tipus i dosis de fertilització.

Informes dels directors

1. Informe dels directors del factor d’impacte de les publicacions presentades

El Dr. F. Xavier Sans i Serra i el Dr. José Manuel Blanco Moreno, directors de la Tesi doctoral de la Roser Rotchés Ribalta, presenten el següent informe sobre el factor d’impacte de les publicacions que formen part de la present memòria.

Capítol 1

Both farming practices and landscape characteristics determine the diversity of characteristic and rare arable weeds in organically managed fields

Rotchés-Ribalta, R., Blanco-Moreno, J.M., Armengot, L., Chamorro, L. & Sans, F.X.

Applied Vegetation Science (2014), doi: 10.1111/avsc.12154

Factor d’impacte (2013): 2.416

Posició dins l’àrea:	Forestry 7/64 (primer quartil)
	Ecology 58/141 (segon quartil)
	Plant sciences 55/199 (segon quartil)

Capítol 2

Which conditions determine the presence of rare weeds in arable fields?

Rotchés-Ribalta, R., Blanco-Moreno, J.M., Armengot, L., José-María, L. & Sans, F.X.

Agriculture, Ecosystems and Environment 203 (2015) 55-61.

Factor d’impacte (2013): 3.203

Posició dins l’àrea:	Agriculture, multidisciplinary 1/56 (primer quartil)
	Environmental sciences 38/216 (primer quartil)
	Ecology 38/141 (segon quartil)

Capítol 3

Herbicide impact on the growth and reproduction of characteristic and rare arable weeds of winter cereal fields

Rotchés-Ribalta, R., Boutin, C., Blanco-Moreno, J.M., Carpenter, D. & Sans, F.X.

Ecotoxicology (2015), doi: 10.1007_sl0646-015-1440-x.

Factor d'impacte (2013): 2.500

Posició dins l'àrea: Ecology 54/141 (segon quartil)
Environmental sciences 71/216 (segon quartil)
Toxicology 39/87 (segon quartil)

Capítol 4

Modulation of the competitive relationship between wheat and the characteristic and rare arable species of winter cereal fields by fertilizers

Rotchés-Ribalta, R., Blanco-Moreno, J.M., Armengot, L. & Sans, F.X.

Sotmès per a la seva publicació a *Weed Research*.

Factor d'impacte (2013): 2.015

Posició dins l'àrea: Agronomy 18/79 (primer quartil)
Plant sciences 67/199 (segon quartil)

Tots els articles que formen part de la Tesi doctoral de la Roser Rotchés Ribalta han estat sotmesos per a la seva publicació a revistes científiques d'àmbit internacional que consten al Science Citation Index (SCI).

Barcelona, maig de 2015

F. Xavier Sans Serra
Departament de Biologia Vegetal
Universitat de Barcelona

José Manuel Blanco Moreno
Departament de Biologia Vegetal
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2. Informe dels directors de la participació de la doctoranda en les publicacions

El Dr. F. Xavier Sans i Serra i el Dr. José Manuel Blanco Moreno, directors de la Tesi doctoral de la Roser Rotchés Ribalta, presenten el següent informe sobre la contribució de la doctoranda en cadascuna de les publicacions presentades en la present memòria.

Capítol 1

Both farming practices and landscape characteristics determine the diversity of characteristic and rare arable weeds in organically managed fields

Rotchés-Ribalta, R., Blanco-Moreno, J.M., Armengot, L., Chamorro, L. & Sans, F.X.

La doctoranda va participar activament en el disseny experimental de l'estudi, en l'execució dels inventaris florístics a camp i en la realització de les enquestes als agricultors. També va dur a terme l'elaboració i l'anàlisi de les dades, la redacció de la primera versió del manuscrit i les revisions posteriors.

Capítol 2

Which conditions determine the presence of rare weeds in arable fields?

Rotchés-Ribalta, R., Blanco-Moreno, J.M., Armengot, L., José-María, L. & Sans, F.X.

La Roser Rotchés Ribalta va participar activament en el disseny experimental de l'estudi, en l'execució dels inventaris florístics a camp i en la realització de les enquestes als agricultors. També va dur a terme l'elaboració i l'anàlisi de les dades, la redacció de la primera versió del manuscrit i les revisions posteriors.

Capítol 3

Herbicide impact on the growth and reproduction of characteristic and rare arable weeds of winter cereal fields

Rotchés-Ribalta, R., Boutin, C., Blanco-Moreno, J.M., Carpenter, D. & Sans, F.X.

La doctoranda va participar en el disseny experimental de l'estudi i en la posada a punt de l'experiment, així com de la recollida de les dades durant l'experiment i de la recol·lecció i processat posterior del material. També va dur a terme l'elaboració i l'anàlisi de les dades, la redacció de la primera versió del manuscrit i les revisions posteriors.

Capítol 4

Fertilization modulating the competitive relationship between wheat and characteristic and rare arable species of winter cereal fields

Rotchés-Ribalta, R., Blanco-Moreno, J.M., Armengot, L. & Sans, F.X.

La doctoranda va participar activament en el disseny experimental de l'estudi i en la posada a punt de l'experiment. També va dur a terme l'elaboració i l'anàlisi de les dades, la redacció de la primera versió del manuscrit i les revisions posteriors.

Finalment, certifiquem que cap dels coautors dels articles abans esmentats ha utilitzat de manera implícita o explícita aquests treballs per a l'elaboració d'una altra tesi doctoral.

Barcelona, maig de 2015

F. Xavier Sans Serra	José Manuel Blanco Moreno
Departament de Biologia Vegetal	Departament de Biologia Vegetal
Universitat de Barcelona	Universitat de Barcelona

Discussió general

Diversos estudis indiquen que la intensificació de la gestió agrícola a escala local i de paisatge ha afectat considerablement les poblacions d'espècies arvenses característiques dels hàbitats cerealistes de secà provocant la seva davallada als sistemes agrícoles i desplaçant-les a les vores dels camps, on avui en dia es troben principalment (Romero et al. 2008b; José-María et al. 2010). No obstant, sovint s'ha posat de manifest la mancança de coneixements específics dels efectes de la intensificació agrícola sobre les espècies segetals a causa de la seva baixa freqüència i abundància. En aquest estudi s'ha superat aquesta limitació mitjançant la realització d'un gran nombre d'inventaris (1957) de la flora segetal a les vores dels camps sota gestió ecològica (304), el que ha permès maximitzar la seva detecció. Aquest mostratge no recull tot el gradient d'intensificació agrícola, atès que ja ha estat descrit en estudis previs que aquestes espècies tenen freqüències molt baixes en camps de gestió convencional (Romero et al. 2008b; José-María et al. 2010). Per tant, es tracta d'una decisió a priori que va permetre centrar l'estudi en els camps sotmesos a una gestió més favorable per a la presència d'aquestes espècies segetals. Això ha permès millorar el coneixement de l'estat de les poblacions d'espècies segetals als secans de Catalunya i relacionar-lo amb les pràctiques agrícoles i amb l'estructura del paisatge. El coneixement dels factors, tant de les pràctiques agrícoles com de les característiques físiques del camp i de la finca, que expliquen la presència d'aquestes espècies és essencial per valorar el seu paper com a indicadors d'una gestió agrícola sostenible enfocada a la conservació de la biodiversitat, especialment de les espècies que han esdevingut rares (*Capítols 1* i *2*).

Els efectes de pràctiques agrícoles concretes sobre l'eficàcia biològica de les espècies segetals també s'han tractat experimentalment. Hem aprofundit en el paper que poden haver tingut l'aplicació d'herbicides i de fertilitzants, com a exemples de pràctiques agrícoles molt comunes, en la disminució de la flora segetal i en determinar la raresa d'algunes d'aquestes espècies (*Capítols 3* i *4*). El coneixement adquirit amb aquests estudis ha de permetre desenvolupar les bases per a propostes encaminades a harmonitzar la producció agrícola i la conservació de la diversitat de la flora segetal.

1. Les espècies segetals als secans de Catalunya

L’estudi de la flora segetal als conreus de la Depressió Central Catalana ha permès detectar més espècies segetals que altres estudis a la mateixa àrea (Romero et al. 2008b; José-María et al. 2010). No obstant, la majoria d’aquestes espècies han estat trobades molt poc freqüentment, fins i tot moltes de les espècies que no són considerades rares per de Bolòs et al. (2005). Amb aquest estudi es va inventariar gairebé el 70 % de les espècies segetals rares citades a la zona estudiada, però moltes d’elles van ser presents en menys de l’1 % de les mostres. Aquests resultats confirmen l’actual raresa d’aquestes espècies, com ja s’havia indicat prèviament (Chamorro et al. 2007), però a la vegada dóna una certa esperança a la possibilitat de la seva conservació ja que moltes d’elles encara són presents als camps de Catalunya.

1.1. Els efectes del paisatge circumdant i de les característiques físiques dels camps sobre les espècies segetals

Diversos estudis assenyalen que els camps immersos en paisatges més complexes solen allotjar una diversitat d’espècies segetals més elevada (Roschewitz et al. 2005; Gabriel et al. 2005) pel fet que els hàbitats adjacents als camps representen hàbitats alternatius que poden actuar com a font de propàguls per a la recolonització dels camps. Aquesta tendència coincideix amb la diversitat d’espècies segetals en conjunt en els camps cerealistes de Catalunya però no amb el subconjunt d’espècies segetals rares, amb les quals el percentatge de terra cultivable hi correlaciona positivament (*Capítols 1* i *2*). La diferència en el patró de resposta de les espècies segetals rares respecte a tot el conjunt d’espècies segetals indica que els hàbitats adjacents no beneficien les espècies segetals rares actuant com a font de propàguls, ja que probablement són espècies més especialistes de l’hàbitat estudiat que les espècies segetals en conjunt (Meyer et al. 2015).

Però l’estructura del paisatge també inclou altres característiques a escala de finca que sí que afecten la diversitat d’espècies segetals. La distribució espacial dels camps d’una finca (distància mitjana entre els camps d’una finca) determina més heterogeneïtat florística entre aquests camps (diversitat β), ja que impedeix l’intercanvi de diàspores. Ara bé, l’estructura de la finca no afecta la diversitat global o γ (*Capítol 1*), de manera que la riquesa d’espècies segetals total a escala de localitat és determinada, principalment, per les condicions que es donen en els camps en si i no tant per l’estructura de la finca o pel paisatge circumdant.

Altres processos d’estructuració del paisatge que afecten la flora arvense, com les característiques físiques del camp, també tenen un efecte significatiu sobre la flora segetal. Els camps més grans, segurament provinents de l’amalgamació històrica de camps més petits, presenten una flora segetal més rica (*Capítol 1*), així com una probabilitat de presència d’espècies rares més alta (*Capítol 2*) que camps més petits, ja que la unió de camps incrementa el conjunt local d’espècies (Marshall 2009). A més, els camps més grans presenten un ventall més ampli de micro-ambients entre els quals es pot donar més fàcilment un intercanvi de diàspores. En canvi, hem hagut de rebutjar la hipòtesi inicial segons la qual esperaríem més diversitat en camps amb formes més complexes, amb més longitud de vora en relació amb l’àrea total, pel fet que aquestes espècies prosperen favorablement en aquestes zones. És possible que aquest resultat reflecteixi la importància de l’àrea en la configuració de la flora local.

A una escala encara més petita, l’efecte de la vegetació present en els marges sobre les espècies segetals de les vores dels camps no és gaire important. Les espècies segetals rares són propenses a ser afectades negativament per la vegetació d’hàbitats adjacents on hi predominen espècies més competitives, com són els boscos i els matollars (*Capítol 2*). La resta d’hàbitats considerats en el nostre mostratge no sembla tenir un efecte diferencial sobre les espècies segetals. Aquests resultats indiquen, doncs, que les espècies segetals responen més a les condicions locals que depenen del camp en si que als hàbitats adjacents com a font de propàguls.

1.2. Les espècies segetals i la gestió agrícola

La gestió ecològica dels conreus cerealistes té efectes positius sobre la diversitat d’espècies arvenses i aquests efectes són acumulatius al llarg del temps. Per aquesta raó, de manera general, els camps i les finques sotmesos a una gestió ecològica durant un període més llarg presenten diversitats d’espècies segetals més elevades (*Capítol 1*). No obstant, la intensitat de les pràctiques agrícoles en els camps de gestió ecològica són molt variables (Clough et al. 2007; Armengot et al. 2011), el que comporta una gran versatilitat dels efectes de l’agricultura ecològica sobre la diversitat vegetal. Per aquest motiu és essencial conèixer els efectes que tenen les pràctiques agrícoles específiques sobre les espècies segetals. Les pràctiques agrícoles incideixen, primordialment, a escala de camp. No obstant, certes pràctiques operen simultàniament i de la mateixa manera sobre tots els camps d'una finca, el que suposa uns efectes sobre la diversitat total i sobre l'heterogeneïtat de les flores entre camps d'una finca (*Capítol 1*).

Les espècies segetals són principalment espècies anuals la persistència de les quals depèn del banc de llavors i de la capacitat de germinar, establir-se i reomplir de nou el banc de llavors. Es tracta d'espècies que germinen, de manera predominant, a la tardor aprofitant la sembra dels cultius, típicament cereals, i completen el seu cicle durant el desenvolupament d'aquests cultius. El notable acoblament dels cicles biològics de les espècies segetals amb l'estacionalitat de les feines agrícoles associades als cereals d'hivern explica la importància del tipus de cultiu per al manteniment de les poblacions d'espècies segetals. Per aquesta raó, el cultiu estant i la gestió associada a aquest cultiu representen uns filtres molt importants de la presència d'espècies segetals (Marshall 2009; Fried et al. 2009).

Les pertorbacions dels hàbitats agrícoles són fonamentals per a l'establiment de les espècies segetals, que són afectades tant per la intensitat com per la naturalesa de les pertorbacions (Critchley et al. 2006). La llaurada representa la pertorbació més important dels agrosistemes arables. Típicament la llaurada en aquests sistemes té lloc a finals d'estiu o durant la tardor, prèvia a la sembra del cultiu. Aquest esquema de pertorbació coincideix amb els requeriments de germinació de la majoria d'aquestes espècies característiques dels sistemes cerealistes de secà (Saatkamp et al. 2011). Per aquesta raó, la sembra de tardor i especialment si és de cereal genera les condicions que afavoreixen la presència d'espècies segetals i rares (*Capítols 1* i *2*). La intensitat de la llaurada també afecta de manera significativa l'establiment de les espècies segetals. Quan es llaura el sòl sense modificar-ne l'estructura vertical (mitjançant l'arada de cisells o la conreadora, per exemple) s'afavoreix la presència d'espècies segetals i rares ja que es generen unes condicions favorables per a la germinació de les llavors que formen part de les capes més superficials del banc de llavors o que hi arriben per dispersió. En canvi, la llaurada que inverteix les capes del sòl (com l'arada de pales) promou l'enterrament de les llavors cap a les capes més profundes del sòl, el que dificulta la germinació i, per tant, l'establiment de nous individus (Gruber & Claupein 2009). Altres fonts de pertorbació, com per exemple la pastura del rostoll, tenen un efecte negatiu sobre la diversitat d'espècies segetals (*Capítol 1*). Encara que aquestes espècies estan adaptades a les pertorbacions periòdiques del sòl, sembla que no toleren bé la pertorbació causada pels herbívors.

Així doncs, els cultius de cereals, als quals les espècies segetals estan adaptades (Kolářová et al. 2013), afavoreixen, d'una banda, la diversitat de la flora segetal que s'hi desenvolupa (*Capítol 1*) i, d'altra banda, la presència d'espècies arvenses rares (*Capítols 1* i *2*). Atès que gran part de les espècies que germinen i prosperen en un any determinat provenen del banc de llavors del sòl, les rotacions que inclouen una proporció de cultius de cereals més elevada determinen més entrades de llavors d'espècies segetals al banc de llavors del sòl, el que propicia el manteniment de les poblacions d'aquestes espècies. El fet que els camps de cereals allotgin més espècies segetals comporta que, en general, les finques que sembren simultàniament més camps de cereals sustenten més diversitat global que aquelles finques amb més diversitat de cultius. En aquestes condicions, els camps són més rics però allotgen una flora més homogènia (la diversitat β és més baixa en finques amb més proporció de camps de cereals) segurament a causa de l'intercanvi més freqüent de llavors amb la maquinària agrícola (Armengot et al. 2012). L'heterogeneïtat de la flora entre camps d'una finca incrementa en relació amb la diversitat de gestió entre camps, condició típica de les finques que també tenen ramaderia. No obstant, aquesta heterogeneïtat florística entre camps no compensa la disminució de la riquesa global de la finca o localitat per la diversitat més baixa dels camps individuals. Per aquest motiu, convé invertir més esforços en promoure i conservar la diversitat de la flora segetal i rara a nivell de camp.

Més enllà de l'establiment, la persistència de les espècies segetals depèn de la seva capacitat de prosperar en un entorn dominat pel cultiu. Les espècies arvenses i el cultiu competeixen pels mateixos recursos i, per tant, tendeixen a ser excloents (González Ponce 1998). Les espècies arvenses característiques dels hàbitats cerealistes de secà són típicament competidores més pobres que els cultius (Critchley et al. 2006) i per això són afectades negativament per les pràctiques agrícoles que afavoreixen el creixement del cultiu. La fertilització dels camps representa un determinant important de les relacions de competència entre el cultiu i les espècies arvenses (Goldberg & Miller 1990; Iqbal & Wright 1997; Tang et al. 2014). L'aplicació abundant de fertilitzants incrementa la disponibilitat de nutrients al sòl per les plantes. Malgrat això, les plantes cultivades són normalment més capaces d’aprofitar els recursos ràpidament i créixer, ocupant de manera eficient l’espai durant l’establiment i disminuint la penetració de la llum (Goldberg & Miller 1990; Robinson & Sutherland 2002; Tang et al. 2014). Aquest fet redueix les possibilitats de creixement de les espècies arvenses característiques i rares dels hàbitats cerealistes de secà (*Capítol 4*). Aquest efecte negatiu de l’elevada aplicació de fertilitzants és força general en les espècies segetals. Per això aquestes espècies són menys freqüents i la diversitat dels camps més baixa en els camps cerealistes de la Catalunya central amb més entrades de fertilitzants orgànics (*Capítols 1* i *2*). Malgrat això, la fertilització orgànica, que allibera els nutrients més lentament que la fertilització mineral, en general afavoreix el desenvolupament de les espècies característiques i de les que són rares dels hàbitats cerealistes de secà. Els efectes negatius de la competència del cultiu són esmorteïts en aquestes condicions, ja que el cereal no creix tan vigorosament com amb la fertilització mineral (*Capítol 4*). Per tant, la fertilització orgànica en quantitats adequades seria una bona estratègia per promoure la conservació de les espècies segetals, alhora que, encara que reduïda, no compromet la producció del cereal.

Les vores dels camps es caracteritzen per unes entrades de fertilitzants més escasses, per haver-hi més errors de sembra i un menor establiment de les llavors del cultiu, pel que el recobriment del cultiu és, en general, baix. Ara bé, quan les condicions són localment favorables, aquestes permeten un bon desenvolupament tant del cultiu com de les espècies arvenses característiques i rares dels hàbitats cerealistes de secà (*Capítols 1* i *2*). Això fa que en les condicions del mostratge trobem una correlació positiva entre la presència d’aquestes espècies i el recobriment del cereal. Aquesta correlació entre el cultiu i la presència d’espècies segetals es manifesta en la variabilitat que trobem dins dels camps, entre punts de mostreig: malgrat que el camp és la unitat bàsica de gestió agrícola, la impossibilitat de portar a terme les operacions agrícoles de manera absolutament homogènia causa certa variabilitat en el recobriment del cultiu (relacionat amb la sembra i l’establiment) i el recobriment total de males herbes. Per tant, a nivell local, quan el cereal s’estableix i prospera adequadament a causa d’unes condicions particularment favorables les espècies segetals rares també se’n beneficien (*Capítol 2*), contràriament a les expectatives inicials.

2. Resposta de les espècies segetals als fertilitzants i als herbicides

Moltes de les espècies segetals han desaparegut dels camps de gestió convencional per efecte de la intensificació de les pràctiques agrícoles que caracteritzen aquests sistemes (Romero et al. 2008b; José-María et al. 2010). L'aplicació d'herbicides ha estat considerada una de les causes principals de la disminució de la diversitat d'espècies en els paisatges agrícoles (Hyvönen & Salonen 2002; José-María et al. 2011). Els herbicides fins i tot poden afectar la vegetació que conforma els hàbitats adjacents, on hi arriben per deriva (Riemens et al. 2008; Dalton & Boutin 2010; Bassa et al. 2011; Schmitz et al. 2014). L'elevada especificitat d'hàbitat de les espècies segetals i la continuada pressió dels herbicides en els sistemes agrícoles de gestió convencional poden haver afectat negativament les seves poblacions. D'altra banda, l'aplicació continuada d'elevades quantitats de fertilitzants minerals també pot haver representat un factor clau en la disminució de la diversitat mitjançant l'alteració de les relacions de competència entre el cultiu i les espècies arvenses (Goldberg & Miller 1990; Iqbal & Wright 1997; Tang et al. 2014).

El tribenuron i el 2,4-D, dos herbicides d'ús comú en els camps de cereals de Catalunya, produeixen importants efectes sobre la biomassa i la reproducció de les espècies segetals avaluades, fins i tot a dosis baixes d'aplicació (*Capítol 3*). Els efectes dels herbicides depenen de la dosi d'aplicació i del tipus d'herbicida aplicat (Clark et al. 2004; Carpenter & Boutin 2010) i, a més, hi ha força variabilitat de resposta entre espècies. Algunes de les espècies segetals testades presenten certa capacitat de recuperació en funció de l'herbicida i de la dosi d'aplicació, ja descrita anteriorment (Riemens et al. 2009; Carpenter et al. 2013). Així, aquelles espècies més afectades o de recuperació més lenta es veuran desplaçades per aquelles més tolerants, resistents o de recuperació més ràpida (Carpenter & Boutin 2010; Carpenter et al. 2013; Egan et al. 2014) o simplement seran excloses competitivament pel cultiu (Christensen 1994; Williams et al. 2008). Per tant, els efectes dels herbicides s'han de considerar dins d'un context de comunitat.

Les aportacions més elevades de fertilitzants minerals no representen un problema per al creixement de les espècies segetals quan creixen soles, el que indica que l'efecte negatiu de la fertilització sobre les espècies segetals té lloc a través de la competència amb el cultiu (*Capítol 4*). D'aquesta manera, el cereal, el qual és més eficient en la captació i l'ús de nutrients, desplaça competitivament les espècies segetals (Robinson & Sutherland 2002). Contràriament, la fertilització orgànica, que allibera els nutrients més lentament, redueix els efectes negatius del cultiu sobre les espècies arvenses característiques i rares dels hàbitats cerealistes de secà.

Atès que la majoria d’espècies segetals són espècies anuals, les pràctiques agrícoles que repercuteixen en la reproducció de les espècies segetals condicionen de manera decisiva la capacitat d’aquestes espècies per prosperar. Tant l’aplicació d’herbicides com la fertilització dels camps poden comportar notables efectes negatius sobre la floració o la formació dels fruits, el que pot reduir la producció de llavors, amb els conseqüents impactes en la reposició del banc de llavors (Strandberg et al. 2012; Qian et al. 2014). Les espècies segetals sotmeses a dosis subletals d’herbicides en general pateixen reduccions més importants en la producció de llavors que en la biomassa total, com ha estat descrit prèviament (Clark et al. 2004; Carpenter & Boutin 2010; Boutin et al. 2014; EFSA Panel on Plant Protection Products and their Residues 2014). El moment de floració de les espècies segetals, però, no va presentar un patró de resposta homogeni en relació amb la competència amb el cereal sota diferents règims de fertilització. Les fertilitzacions baixes i la fertilització orgànica tendeixen a causar endarreriments de l’inici de la floració, però en pocs casos aquest efecte és estadísticament significatiu. Malgrat tot, això podria tenir importants implicacions per a les poblacions ja que quan les espècies floreixen més tard, solen tenir menys descendència (Storkey 2006; Fried et al. 2012).

Els resultats d’aquests estudis sota condicions controlades són de gran rellevància ja que permeten avaluar l’efecte individual de determinades pràctiques sobre el desenvolupament de les espècies testades. A la vegada, evita la interferència d’altres variables que també poden influir en el comportament de les espècies, el que dificulta la comprensió dels resultats (Dalton & Boutin 2010). A més, són absolutament irreemplaçables per a la quantificació dels efectes de certes pràctiques agrícoles (l’aplicació d’herbicides i de fertilitzants) sobre les espècies segetals i, en particular, per a la detecció dels efectes que tenen lloc sota una agricultura convencional, la més estesa en el territori. Les baixes freqüències de les espècies segetals que es donen actualment en els camps de gestió convencional d’una banda dificulten notablement la detecció d’aquestes espècies i d’altra banda, impossibiliten l’estimació dels efectes amb precisió. Malgrat això, els resultats d’aquest tipus d’aproximacions s’han de prendre amb precaució, sobretot a l'hora d'extrapolar-los al funcionament de les comunitats naturals. La coexistència amb altres espècies, la manca de transitivitat en les relacions de competència, la variabilitat temporal i espacial, entre d'altres, són limitacions lògiques a les que està sotmesa qualsevol experimentació.

3. Factors determinants de raresa de les espècies segetals

L'aplicació d'herbicides ha estat considerada una de les causes principals de la disminució de la diversitat d'espècies en els paisatges agrícoles (Hyvönen & Salonen 2002; José-María et al. 2011). Per aquest motiu s'esperava que aquelles espècies rares o menys freqüents en l'actualitat presentessin una sensibilitat als herbicides més elevada que espècies més comunes de la mateixa família. De la mateixa manera, com que les elevades entrades de fertilitzants als camps de cereals afavoreixen el cultiu en detriment d'aquelles espècies amb menys capacitat competitiva, es va considerar que les espècies arvenses rares estarien en desavantatge sota aquestes condicions (Storkey et al. 2010; Kovács-Hostyánszki et al. 2011). Però contràriament a les hipòtesis inicials, els nostres resultats experimentals no donen suport a la relació entre la raresa i la sensibilitat als herbicides o la capacitat competitiva de les espècies segetals rares en comparació amb les més comunes a diferents tipus i dosis de fertilització (*Capítols 3* i *4*). Per algunes parelles d'espècies testades, la més rara va ser efectivament la més sensible a un determinat herbicida o més afectada per la competència amb el cultiu que les espècies comunes aparellades. Però en altres casos, les comunes van resultar més afectades, o les dues espècies de la mateixa família van mostrar comportaments similars.

Es pot considerar que tant l'aplicació d'herbicides com de fertilitzants a altes dosis han estat factors importants a l'hora de provocar la pèrdua generalitzada de diversitat vegetal arvense. Per tant, convindria potenciar un ús reduït dels herbicides i dels fertilitzants, especialment els minerals, a les vores dels camps de gestió convencional per facilitar la conservació de les espècies segetals. Malgrat això, la sensibilitat als herbicides i la resposta competitiva sota fertilització individualment són uns predictors pobres de la raresa de les espècies segetals. És molt probable, doncs, que les baixes freqüències d'algunes espècies segetals hagin estat causades per diversos factors, o la combinació d'aquests, que han afectat les espècies de manera diferencial.

L’avaluació de la presència de les espècies segetals rares en els camps de cereals de la Catalunya central permet preguntar-se quin és l’abast de la relació entre les pràctiques agrícoles, l’estructura del paisatge i la raresa d’algunes d’aquestes espècies. És difícil establir uns models estadístics sòlids que permetin destriar quines són les condicions i les pràctiques agrícoles que afecten significativament la presència de les espècies segetals rares, ja que malgrat seleccionar les que són més freqüents, aquestes no deixen de ser espècies inusuals. Globalment, però, aquestes espècies presenten uns patrons comuns de resposta a diverses variables considerades en el model (*Capítol 2*). Les condicions que correlacionen negativament amb la diversitat de les espècies arvenses segetals també ho fan amb la probabilitat de trobar les espècies segetals rares, com ha estat descrit prèviament (Romero et al. 2008b). Aquesta tendència comuna per a les espècies segetals rares indica que en general tendeixen a aparèixer de manera preferent sota determinades pràctiques agrícoles. De manera semblant a la diversitat d’espècies segetals, la sembra de cereal, especialment de tardor després d’una llaurada sense inversió del sòl, poden afavorir la conservació d’aquestes espècies en camps agrícoles de gestió ecològica.

Malgrat tot, els patrons que determinen la presència d’espècies segetals rares són menys evidents que per a la riquesa específica. Els factors aleatoris tant a nivell de camp com de localitat sovint tenen més pes sobre la presència de les espècies rares que les condicions locals o les variables de gestió i de paisatge (*Capítol 2*). A part de les variables considerades, la presència d’aquestes espècies depèn, en gran mesura, del camp i de la localitat en si. És a dir, la presència d’espècies segetals rares sembla ser el resultat de la interacció de factors estocàstics i dels factors històrics dels camps i de les localitats, que determinen el conjunt d’espècies local (Hiltbrunner et al. 2008), amb la gestió, que en condiciona l’expressió en un any determinat. Les actuacions de conservació de les espècies segetals, doncs, s’haurien de focalitzar en aquells camps on aquestes espècies han estat trobades per evitar la seva pèrdua.

4. Implicacions per a la conservació

Les espècies segetals són un dels grups que més indiscutiblement ha experimentat una intensa regressió a Catalunya (Chamorro et al. 2007) i que, a hores d’ara, plantegen més problemes de conservació en gran part d’Europa (Sáez et al. 2011). La repetida aplicació de diferents herbicides als camps de cereals per al control de les espècies arvenses ha estat una de les causes principals de la disminució de la diversitat vegetal, ja que provoquen efectes importants sobre el creixement i la reproducció de les espècies segetals. D’altra banda, la fertilització també ha repercutit sobre aquestes espècies de manera indirecta, ja que afavoreix les espècies cultivades (i probablement les espècies més competitives) en detriment de les espècies segetals. Ara bé, la freqüència especialment baixa d’aquelles espècies que es consideren rares sembla ser la conseqüència de diversos factors, segurament de la interacció entre ells, i sempre amb una incidència que varia d’una espècie a una altra. Per tant, la promoció de mesures de gestió encaminades a la conservació d’aquelles espècies més rares requereix estudis específics que permetin discernir les pràctiques més influents en cada cas.

La gestió ecològica sembla una bona mesura per afavorir les espècies segetals, que encara són presents a les vores dels camps. Malgrat tot, són espècies molt poc freqüents fins i tot als camps gestionats de manera ecològica de la regió cerealista de la Catalunya central. Per aquest motiu considerem que ara és el moment d’emprendre mesures per a la seva conservació. Els fonaments per a la conservació de les espècies segetals han d’anar més enllà de la gestió ecològica ja que hi ha determinades pràctiques de la gestió ecològica dels cultius cerealistes que no beneficien les espècies segetals. S’haurien de promoure les pràctiques agrícoles específiques que potencien la presència d’espècies segetals, tals com la sembra de cereal, especialment de tardor, després de llaurades sense inversió de les capes del sòl. A la vegada, però, convindria racionalitzar la fertilització, encara que aquesta sigui orgànica, per no comprometre la producció i, alhora, garantir la persistència d’aquestes espècies. D’altra banda, caldria restringir la pastura en aquelles zones on s’han trobat algunes espècies segetals, sobretot si es consideren rares.

Altres accions adreçades a nivell de finca o de paisatge són menys factibles i tenen menys probabilitat de ser eficaces. D’una banda, els factors que actuen a escala local tenen uns efectes més importants sobre la presència d’espècies segetals i, d’altra banda, les condicions a les escales de finca i de paisatge són menys propenses a ser modificades. No obstant, s’hauria de promoure la producció de cereals a nivell de finca, especialment en finques amb els camps allunyats, per tal de desenvolupar estratègies de conservació eficaces que afavoreixin la presència d’espècies segetals així com l’intercanvi de diàspores i l’establiment d’espècies segetals en camps nous.

Conclusions generals

En aquesta tesi s'ha estudiat la freqüència i la diversitat de les espècies arvenses característiques dels hàbitats cerealistes de secà, o espècies segetals, a les vores de camps de gestió ecològica de la Catalunya central. S'ha avaluat com depenen de les característiques del paisatge que envolta els camps, de la gestió dels camps i de les condicions locals dins els camps. S'ha determinat l'abast d'aquests efectes per al conjunt d'espècies segetals trobades a la zona d'estudi, així com per a les espècies segetals considerades rares. D'altra banda, s'ha analitzat el paper que han pogut tenir certes pràctiques agrícoles habituals, com són l'ús d'herbicides i la fertilització, en determinar les diferències en la freqüència actual d'algunes espècies segetals. Aquests estudis han permès determinar quines pràctiques de gestió agrícola s'haurien de promoure per tal de potenciar la conservació d'aquestes espècies.

A continuació s'exposen breument les principals conclusions dels diversos treballs recollits en aquesta tesi doctoral.

- Les espècies segetals que han estat citades a Catalunya apareixen en freqüències molt baixes, fins i tot en els camps de gestió ecològica. Aquest fet corrobora el seu estat delicat de conservació i indica la necessitat de promoure estratègies per a la seva preservació ara que encara moltes d'elles hi són presents.

- De manera general, malgrat que la continuïtat de la gestió ecològica afavoreix la diversitat d'espècies segetals, els efectes de les diferents pràctiques agrícoles usades són força variables. Per tant, convé avaluar individualment les pràctiques agrícoles que poden afectar les poblacions d'espècies segetals per desenvolupar les estratègies de conservació.

- Les pràctiques agrícoles a nivell de camp representen uns determinants importants de la riquesa d'espècies segetals i rares. La sembra de cereal en un camp, preferentment a la tardor després d'una llaurada sense inversió de la capa superficial del sòl, afavoreix el conjunt d'espècies segetals, a la vegada que genera uns ambients favorables per a la presència de la majoria de les espècies segetals rares.

- La fertilització també és un factor que afecta significativament la freqüència d'espècies segetals i rares en els camps cerealistes de secà de Catalunya. L'excessiva disponibilitat de nutrients estimula la competència exercida pel cultiu, en detriment del creixement de les espècies segetals i de les que són rares.

- El percentatge de terra cultivable al voltant de cada camp té un efecte negatiu sobre la diversitat d'espècies segetals que es pot trobar en un camp (diversitat α), però positiu sobre la diversitat de les espècies rares. Aquests resultats contradictoris indiquen que les espècies segetals rares són més especialistes dels hàbitats cerealistes de secà en comparació amb el conjunt global d'espècies segetals.

- L'àrea del camp té un efecte positiu sobre la diversitat d'espècies segetals global i la diversitat d'espècies rares, de manera que camps més grans sostenen més espècies segetals, a la vegada que hi ha més probabilitat de trobar-ne de rares. En canvi, camps amb una relació perímetre/àrea més alta presenten menys espècies segetals.

- La diversitat total d'espècies segetals en una finca (diversitat γ) depèn, en primera instància, de la riquesa específica dels camps (diversitat α) més que no pas de l'heterogeneïtat entre les flores dels camps (diversitat β). Per tant, una proporció més alta de camps amb cereals a la finca, els quals sostenen una flora segetal més rica (diversitat α) però distribuïda més homogèniament entre ells, determina una diversitat d'espècies segetals a la finca (diversitat γ) més elevada.

- Malgrat que la diversitat total de la finca no incrementa, les finques que combinen la ramaderia i l'agricultura tenen una flora més heterogènia entre els camps a causa de la diversificació en la gestió dels diferents camps. Camps més separats entre ells també determinen una flora més heterogènia a la finca.

- Malgrat és difícil trobar models estadísticament significatius que expliquin la presència de les espècies segetals rares, existeixen patrons de resposta semblants entre espècies segetals rares davant de les característiques del paisatge, de la gestió i de les condicions locals avaluades.

- Les espècies segetals rares apareixen més freqüentment on hi ha més recobriment d'espècies arvenses en general, el que indica que les condicions que afavoreixen l'abundància d'espècies arvenses, també afavoreixen la presència d'aquelles més rares. Les espècies segetals rares tendeixen a evitar localment les vores al costat de marges més competitius tals com boscos o matollars, però no presenten una preferència remarcable per cap altre tipus d'hàbitat dels marges, els quals no actuarien especialment com a refugi.

- Les espècies segetals rares tendeixen a aparèixer a les vores dels camps on el recobriment del cultiu és més alt. És així ja que a les vores dels camps el recobriment del cultiu no sol ser gaire alt i quan es donen unes condicions adequades pel cultiu, també beneficien el creixement de les espècies segetals.

- Els factors estocàstics que depenen de la història particular dels camps i de les finques adquireixen importància a l'hora de determinar la presència d'espècies segetals rares als camps, sovint més que les pràctiques agrícoles.

- L'aplicació d'herbicides afecta les espècies segetals testades fins i tot a dosis baixes. Però la seva sensibilitat als herbicides depèn del tipus d'herbicida, essent les respostes específiques d'espècie.

- En general, els efectes a curt termini dels herbicides sobre la biomassa són més manifestos que a llarg termini, fet que indica certa capacitat de les espècies segetals de recuperar-se. Els patrons de recuperació depenen, en gran part, de la dosi d'aplicació, requerint més temps com més altes són les dosis d'aplicació d'herbicides.

- Els efectes a llarg termini dels herbicides són més acusats sobre la capacitat reproductiva que sobre la biomassa total. Atès que les espècies segetals són plantes anuals que depenen de la reproducció per assegurar la seva persistència, l'anàlisi a curt termini mitjançant la biomassa com a única mesura dels efectes dels herbicides no és un criteri adequat per a l'avaluació d'aquests efectes sobre les poblacions de les espècies segetals.

- Les diferències en la sensibilitat als herbicides entre espècies segetals filogenèticament properes no poden explicar de manera general les diferències en la freqüència actual entre aquestes espècies.

- Les espècies segetals avaluades no manifesten problemes per créixer en condicions de fertilització mineral alta, però aquesta fertilització intensifica els efectes negatius que té la competència del cereal sobre el creixement d'aquestes plantes en comparació amb la fertilització orgànica. Per tant, la fertilització orgànica seria una bona estratègia per a promoure el desenvolupament de les espècies segetals a la vegada que no compromet gaire la producció del cultiu.

- No totes les espècies segetals rares responen de la mateixa manera en comparació amb l’espècie comuna davant la pressió de competència del cereal sota diferents tipus i dosis de fertilitzants.

- Malgrat els efectes dels herbicides i de la fertilització han contribuït clarament en l’enrariment de les espècies segetals, els nostres resultats indiquen que les diferències en els seus efectes sobre les espècies particulars no correlacionen amb la seva freqüència al territori. Les causes de la raresa d’espècies segetals particulars s’han de cercar en altres factors o en la combinació de diferents factors.

Effect of farming practices and landscape on the segetal species of Mediterranean dry land arable fields

Implications for conservation

English version

General introduction

1. Agricultural systems

1.1. General aspects

Agricultural systems are man-made systems. Their origin and maintenance depend on the human intervention, which replaced natural habitats mainly to produce food. The antiquity and the nature of that intervention have led to an assembly between agricultural practices and the resulting semi-natural ecosystems. For this reason, agriculture has a key role not only in the food supply but also in the maintenance of the whole agricultural ecosystems.

Agricultural land occupies an important part of the landscape, being 37.6 % of the land area, which places it as the main land use worldwide in terms of extension (Food and Agriculture Organization of the United Nations 2013). The proportion is similar in Catalonia, where the area devoted to crop production is 1 125 268 ha, the 35.1 % of the land (Institut d’Estadística de Catalunya 2013). Almost 70 % of this area is dedicated to the production of arable crops, from which more than two thirds are rain-fed (Institut d’Estadística de Catalunya 2013). Over the 30 % of the agricultural land area of Catalonia (380 648 ha) is devoted to cereal crop production. Given their extension, both worldwide and regionally, these habitats are of great importance for land conservation.

1.2. Agricultural intensification

Agricultural management has changed in relation to technological innovation to meet the food demands of an increasing world population. These changes became more noticeable from the mid twentieth century with the so-called ‘Green Revolution’, with which the world cereal production doubled in only 40 years (Tilman et al. 2002). Available data of cereal production in Spain also support it (Figure 1). However, this increase in crop production has taken place at the expense of overall environmental quality (Tilman et al. 2001). Both the expansion of agricultural areas at the expense of natural ecosystems and the intensification of management to increase the productivity (Foley et al. 2011) have been key factors to increase the crop production, but they are also the main cause of the environmental impacts. The analysis of the agricultural changes between 1985 and 2005 revealed that a 28 % increase of global food production took place while cropland area increased only 2.4 % (Foley et al. 2011). Therefore, the intensification of farm management, owing to the use of highly productive crop varieties, mechanization of farming, application of chemical fertilizers and the use of pesticides, among others, has been the main cause of the increase of agricultural production (Matson et al. 1997), as shown by the trends in Spain over the last 50 years (Figure 1).

The intensification of agricultural land use to increase its productivity has often taken place at the expense of the ability of agroecosystems to provide goods and services. For instance, high applications of fertilizers and pesticides (Figure 1) reduce the quality of the soil and pollute the groundwater (Tilman et al. 2002).

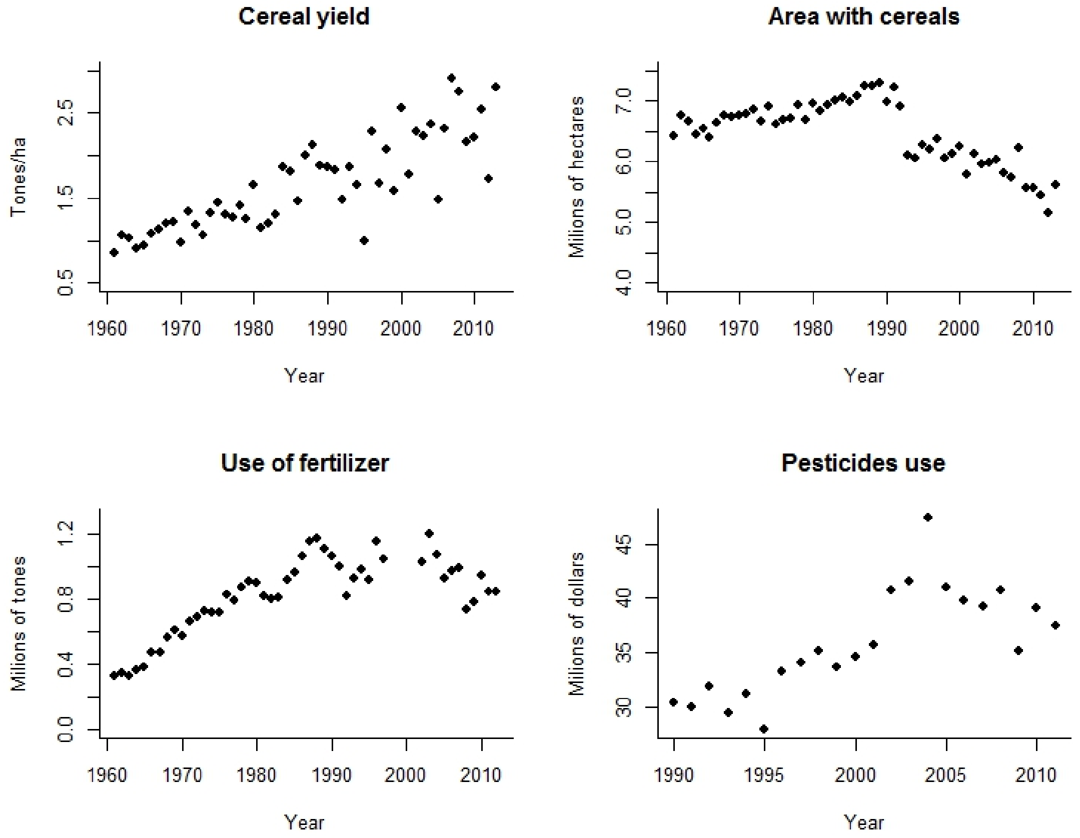

Figure 1: Agricultural trends in Spain over the past 50 years. Cereal crop yield, area with cereals sown, use of fertilizers and overall pesticides use. Data were obtained from Faostat (2015).

The intensification of agriculture also exerted significant changes in species composition of biotic communities and it is the main cause of the dramatic decline of biodiversity (Robinson & Sutherland 2002; Sans et al. 2013). Despite the effects of agricultural intensification are widespread, the involved processes act at specific scales, both at the landscape scale (Robinson & Sutherland 2002; Tscharntke et al. 2005) and at the field scale, where local farm management is very important (Benton et al. 2003; Norton et al. 2009).

1.2.1. Agricultural intensification at a landscape scale

The intensification that has characterised agriculture over the last few decades has involved important changes on the agricultural landscape. Field consolidation during the second half of the twentieth century led to field aggregation and to a structural simplification of many habitats on the field boundaries and other plant formations interspersed in agricultural land (Stoate et al. 2009). Thereby, agricultural landscape in some areas has suffered a simplification process: from complex landscapes consisting of a mosaic of crops and boundaries and surrounded by a high proportion of natural and semi-natural habitats to simple landscapes, dominated by large extensions of cultivated land with little structural heterogeneity (Robinson & Sutherland 2002; Roschewitz et al. 2005; Tscharntke et al. 2005; Baessler & Klotz 2006). The loss of habitats at the adjacent boundaries and the fragmentation of natural habitats because of expanding agriculture have increased the extinction risk of species with small and isolated populations, which has resulted in the loss of diversity (Roschewitz et al. 2005; Tscharntke et al. 2005).

1.2.2. Agricultural intensification at the field scale: agricultural management

Agricultural intensification at the field scale is mainly an outcome of field management. The management that has characterised the agriculture of developed and developing countries over the last decades constitutes what is known as conventional agriculture (Gliessman 2000). In general, it relies on high inputs of organic and inorganic fertilizers and chemical pesticides, the mechanisation of farm management which allowed intensive tillage, the simplification of crop rotation, the sowing of large extensions of monocultures and the use of commercial seeds free from weeds, among others. These practices affect the environmental quality and biodiversity at field and farm scales (Stoate et al. 2009) but may also have negative effects on the surroundings of the farms (Matson et al. 1997; Robinson & Sutherland 2002; Tilman et al. 2002; Foley et al. 2011).

To mitigate the effects of farming practices, different environmentally-friendly management strategies focused on sustainability have emerged. These farming strategies integrate the experience of traditional agriculture together with scientific and technological knowledge with the aim to decrease the negative environmental impacts of the conventional agriculture (Matson et al. 1997). These methods pretend to preserve soil fertility and quality while promoting the biodiversity and preserving the natural ecological balance (Food and Agriculture Organization of the United Nations 2002). They represent, in a certain way, a return to traditional management; they base the fertilization on the application of organic matter and on the inclusion of legumes and green manures in the crop rotation and on a mechanical control of the weeds, among other practices. Although it is a good strategy to preserve the habitat quality, organic management does not reach yields as high as conventional agriculture (Food and Agriculture Organization of the United Nations 2002; Seufert et al. 2012). Probably because of this among other reasons (Musshoff & Hirschauer 2008), organic management is still a productive model of minor importance, which only represents the 2 % of the agricultural land in Europe and less than the 10 % in Catalonia. However, the area occupied by organic farming has increased considerably over the last years. For instance, from 2003 to 2013 the agricultural area managed organically has doubled in Catalonia (Consell Català de Producció Agrària Ecològica 2013).

1.2.3. Effects of agricultural intensification on biodiversity

One of the most significant effects of agricultural intensification both at the field and landscape scales has been the loss of biodiversity (McLaughlin & Mineau 1995; Robinson & Sutherland 2002; Tilman et al. 2002). Agricultural systems, understood as the mosaic of cultivated land and natural and semi-natural habitats, encompass a large proportion of biodiversity in relation to other terrestrial habitats (Pimentel et al. 1992). Nevertheless, these systems have suffered larger losses over the last few decades (Preston et al. 2003) (Figure 2). Therefore, it is worth investing efforts on the conservation of the species inhabiting agricultural areas that have been more negatively affected by agricultural intensification (Pimentel et al. 1992). It is actually a complex balance as it entails the need to harmonize agricultural production and the maintenance of the diversity of many biological groups inhabiting agroecosystems (Sans 2007; Zhang et al. 2007).

Although we generally talk about biodiversity loss, changes in the species composition are also noticeable. While there is a trend for many species to decrease their frequency, few find suitable conditions and become dominant. In addition, these more abundant species are often generalist species, which leads to the banalisation of these communities (Albrecht 2003). Therefore, the most remarkable aspect of this biodiversity loss within agricultural habitats has been the loss of more specialised taxa, which are found preferentially in low intensified farming systems (Robinson & Sutherland 2002). These effects of agricultural intensification have been described on different groups of organisms but particularly birds, arthropods and plants that somehow depend on farming systems (Robinson & Sutherland 2002).

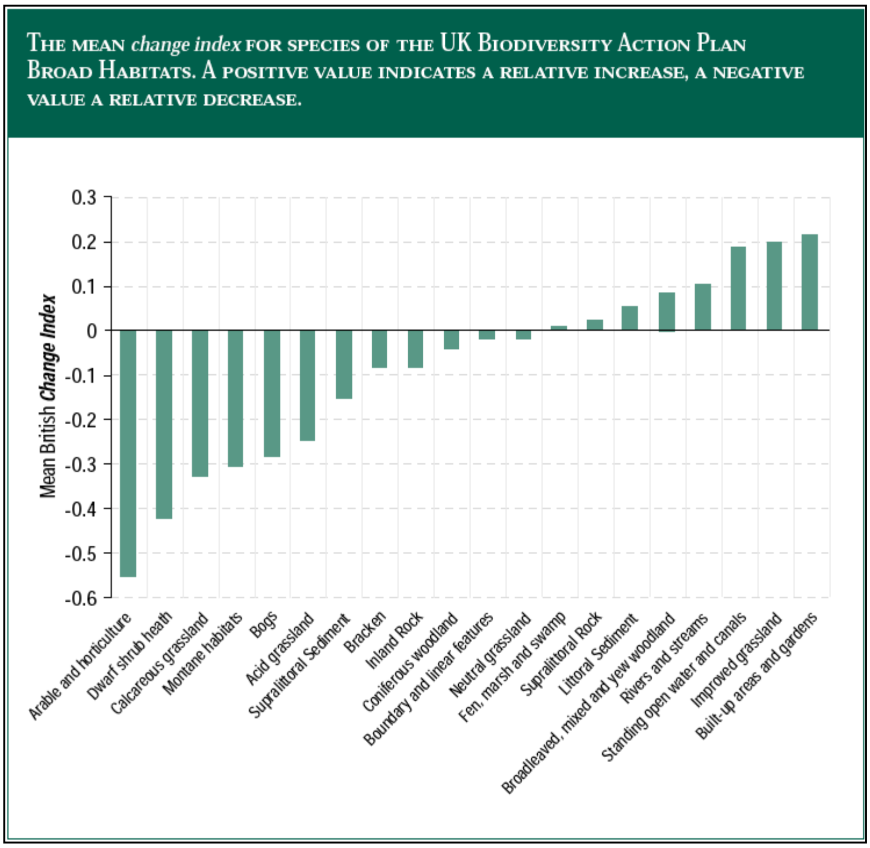

Figure 2: Index of the mean change of species diversity in United Kingdom per habitat. Positive values indicate an increase of biodiversity while negative values show a relative decrease of biodiversity when comparing the surveys from 1930-1969 and 1987-1999 (Preston et al. 2002).

2. Plant diversity of agroecosystems

2.1. Plant diversity associated to agricultural systems

Arable species are the plants that colonize the crops but are not intentionally sown by the farmer. The development and the expansion of agriculture in its origins involved the selection of a flora thriving in these habitats. The arable plant diversity, thus, depends on the establishment of newly dispersed seeds and from the soil seed bank (Ryan et al. 2010). Therefore, arable diversity is determined by the environmental factors such as the weather, the landscape context and the physical characteristics of the soil, but also the farm management (Roschewitz et al. 2005; José-María et al. 2010).

Arable plant species, also known as weeds, have been traditionally viewed as an impediment to crop production as they compete for the same resources. Weeds may cause losses of around the 30 % of the harvest (Oerke 2006). Moreover, arable species can act as a host for some pests and diseases of cultivated plants and, thus, negatively affect both crop yield and quality (Masalles 2008). That is why the reduction of arable weeds populations has been one of the main objectives of the arable farm management. Nevertheless, only few of the many arable species are actually a threat to crop production. Most of them rarely cause large crop yield losses, but considerably contribute to species diversity in arable habitats (Albrecht 2003).

The biodiversity of agroecosystems is an essential part of our natural heritage, and is related to the history of land use. Specifically, it is difficult to understand the Mediterranean landscapes without agriculture. Therefore, arable plant species have an intrinsic value as being part of the biodiversity of these systems. They have important aesthetic and conservation values of agricultural landscapes, and are indicators of a traditional and low intensive agriculture (Aboucaya et al. 2000; Clergue et al. 2005). In addition, arable plant diversity plays a key functional role because they provide alternative resources for pollinators, herbivores and granivorous animals and they are the habitat and refuge for some crop-associated fauna (Altieri 1999; Marshall et al. 2003; Clergue et al. 2005).

2.2. The segetal flora

The segetal flora is the suite of the arable species inhabiting preferentially the dry land cereal farming systems. Some authors have named them as characteristic arable species or obligate arable weeds (Sutcliffe & Kay 2000; Albrecht 2003; Romero et al. 2008; José-María et al. 2010). The segetal flora includes many species from the Middle East, from where they were introduced by humans with the expansion of agriculture, becoming archaeophytes. These are naturalized non-native species that were introduced to a certain region, before 1500 AD (Pyšek et al. 2004). However, the segetal flora also includes some species that, despite being native, they are nowadays mainly found in arable fields.

Most characteristic arable species are annual species that require the periodical disturbances of arable fields to re-establish their populations from the seed bank or from the propagules arriving from the adjacent fields every year. Their persistence often depends on a high fecundity and on the ability to germinate over relatively long periods (Masalles 2008). At the same time, these segetal species are unable to thrive in more competitive habitats of the adjacent boundaries (Marshall 2009). The persistence of their populations in most places, thus, seems to require the maintenance of farming. That is why these species are considered characteristic of the phytosociological order *Secalietalia cerealis* Br.-Bl. 1936, gathering all the weed communities of dry land winter cereal fields.

The assessment of the weed diversity within arable fields have been usually conducted considering all plant species that are not intentionally sown by the farmer (Roschewitz et al. 2005; Hiltbrunner et al. 2008). However, it may be more appropriate to consider only the specific species of dry land cereal fields because they do not include the ones frequently appearing outside the fields. Therefore, the most generalist species such as creeping thistle (*Cirsium arvense* (L.) Scop.) or cleaver (*Galium aparine* L.) were not considered to assess the diversity (Albrecht 2003).

2.2.1. Impact of agricultural intensification on the segetal flora

Despite characteristic arable species are adapted to the periodic disturbances of farm management, the agricultural intensification over the last decades may be the cause of the remarkable decline of their populations (Sans et al. 2013). In general, specialist species of any habitat are more sensitive to changes in the environmental conditions and land use than more generalist species (Albrecht 2003). As a consequence, agricultural intensification has caused a decrease of characteristic arable species populations such as the common corncockle (*Agrostemma githago* L.), cornflower (*Centaurea cyanus* L.), *Hypecoum procumbens* L., corn buttercup (*Ranunculus arvenses* L.) or *Roemeria hybrida* (L.) DC., among others. At the same time, more generalist species have become more abundant (Masalles 2008; Romero et al. 2008b).

The diversity loss of characteristic arable species has been found in different European countries such as Germany (Albrecht 1995), Denmark (Andreasen et al. 1996), Hungary (Kovács-Hostyánszki et al. 2011), United Kingdom (Sutcliffe & Kay 2000; Robinson & Sutherland 2002) and Spain (Cirujeda et al. 2011). This issue has also been addressed in Catalonia (Chamorro et al. 2007; Romero et al. 2008b), although it may not have received the appropriate attention (Sáez et al. 2011).

The changes that have taken place in arable land management over the past 60 years are so many that it is unlikely that any one factor alone can adequately explain the decline in arable farm diversity (Sutcliffe & Kay 2000). However, the simplification of crop rotations (Hyvönen & Salonen 2002) as well as the intensive tillage (Mas & Verdú 2003; Santín-Montanyá et al. 2013) may have had a significant contribution towards the decrease of the weed diversity of dry land arable fields. For some characteristic arable species, the improvement in seed cleaning techniques has been considered the cause of the decline of their populations (Firbank 1988).

However, it is considered that the use of herbicides to control weeds has been one of the farming practices that have most affected arable species populations (Robinson & Sutherland 2002; Potts et al. 2010). Since the introduction of the 2,4-D herbicide in 1946, the development and use of new products has been related to the decrease of arable species diversity (Heap 1997). At the same time, the constant application of herbicides in arable fields has resulted on the rise of resistant species, most of which have become actually harmful (Heap 1997; Kudsk & Streibig 2003; Ulber et al. 2010).

The increased use of fertilizers, especially of those obtained by chemical synthesis, may also have exerted a significant pressure on the characteristic arable species diversity. This increase of the nutrient availability in the soil can cause toxic effects on plants, but more generally it alters the competitive interactions between the crop and the arable weed species in favour of the former (Pyšek & Lepš 1991). Consequently, the segetal flora has become a poorer competitor (Kovács-Hostyánszki et al. 2011). This effect is magnified by the use of highly competitive crop varieties. Organic fertilizers, despite they may provide the same amount of nutrients, their release is generally slower, which reduces the nutrient concentration. Therefore, organic fertilizers may palliate the negative effects of fertilization on arable species diversity (Robinson & Sutherland 2002).

The changes occurred on the agricultural landscape structure have also contributed to the loss of characteristic arable species diversity (Kovács-Hostyánszki et al. 2011; Solé-Senan et al. 2014). Characteristic arable species have been negatively affected by the landscape simplification. Several studies suggested that this simplification process involved, on one hand, the reduction of the extent of natural and semi-natural habitats that could act as a refuge and, on the other hand, the facilitation of the intensification of farm management as a result of homogenization of the landscape.

2.2.2. The distribution of segetal flora

The effects of the farm management and of the landscape structure on the segetal flora have a varying intensity on the boundaries, on the edges and on the centre of the field. Agricultural management has a greater impact at the centre of the field and decreases towards the field edges and boundaries. Contrarily, the effects of the landscape structure are more noticeable at the boundaries and at the field edges but have a minor effect on the diversity of the field centre (José-María et al. 2010; Kovács-Hostyánszki et al. 2011). Thus, the plant diversity is concentrated in the boundaries and decreases towards the field centre. However, characteristic arable flora behaves differently because it concentrates on field edges (Romero et al. 2008b; Fried et al. 2009) (Figure 3).

Field edges are defined as the outermost cultivated meters of the fields adjacent to the boundaries of natural or semi-natural habitats (Marshall & Moonen 2002). At the field edges, agricultural practices are performed less efficiently because of the difficulty of access to the machinery. For this reason, the amount of fertilizers and herbicides received at the edges is generally lower, the intensity of tillage is decreased and there are more sowing failures, which reduces the competitive pressure of the crop on the arable weed species (Kleijn & van der Voort 1997; Dutoit et al. 2007; Romero et al. 2008b).

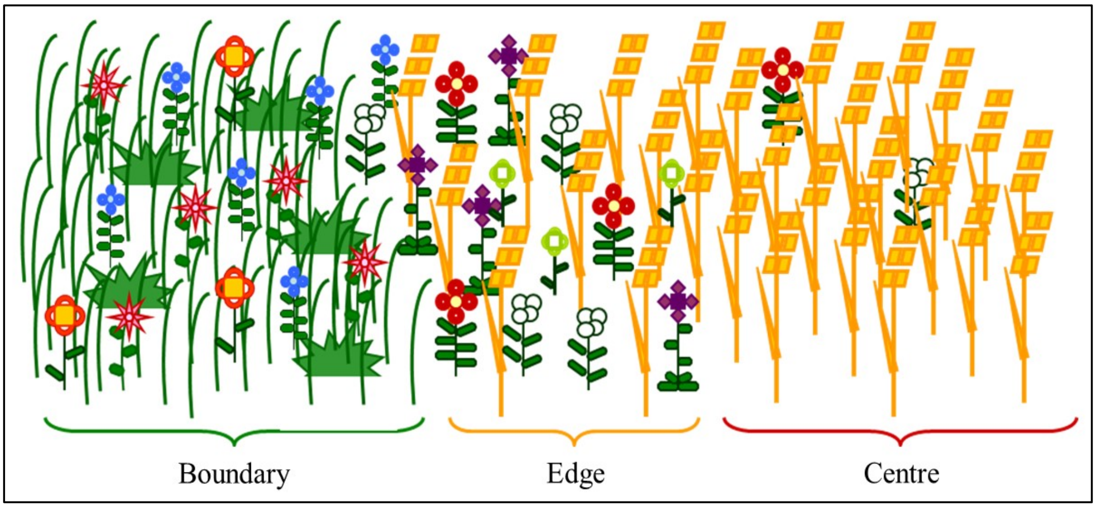

Figure 3: Diagram of the spatial distribution of species in the fields, from the boundary to the field centre. The plant diversity is higher at the boundaries and lower towards the field centre. However, the segetal flora or characteristic arable species of dry land cereal fields inhabit preferentially at the field edges, because these species require the periodic disturbances of the dry land arable fields at the same time that it benefits from the lower efficacy of farming practices such as fertilization and weed control in this area.

2.2.3. The rare segetal species

The major impact of the agricultural intensification of the last decades on the characteristic arable species has been a significant decline of their populations, to the extent that some of them have become rare or even locally extinct (Baessler & Klotz 2006; Fried et al. 2009; Cirujeda et al. 2011; Storkey et al. 2012). Many European countries have considered these less frequent arable species in their red lists (Schnittler & Ludwig 1996; Moser et al. 2002; Cheffings & Farrel 2005; Colling 2005; Türe & Böcük 2008; Moncorps & Siblet 2012). This consideration raises their conservation value, which constitutes the basis for most conservation strategies. In some countries, rare segetal species have been considered as priority species for conservation in action plans such as in the United Kingdom (Preston et al. 2003) or in France (Aboucaya et al. 2000).

Unfortunately, in the Mediterranean region characteristic arable species have attracted relatively little the attention of researchers and conservationists. For instance, rare arable species are not considered in the Red book of endemic and endangered vascular plants of Catalonia (Sáez et al. 2011) because they depend on the maintenance of anthropic systems and some of them are introduced. The only assessment of their frequency in the region has been carried out in the local floras, using a non-systematic approach and in relation to the whole regional flora; there is not a detailed analysis of the status of their populations. Despite the apparent loss of the segetal species in Spain and in Catalonia (Chamorro et al. 2007; Cirujeda et al. 2011) conservation strategies have not been implemented to conserve the arable plant species, or only very locally (Mateu & Segarra 1998).

Agri-environmental schemes (AES) have been put forward at the European level (European Environment Agency 2004), which require to the member states the development and implementation of agro-environmental programs based on financial incentives with the aim to mitigate the negative impact of agricultural intensification. These programs are not only aimed at preserving the biodiversity but also at the maintenance and improvement of the environmental quality. Organic farming has been promoted as an AES measure in some countries (Kleijn et al. 2006); farming systems managed less intensively may promote the characteristic arable species (Kleijn et al. 2006; Rundlöf et al. 2009; Gabriel et al. 2010; Kolářová et al. 2013). However, the effectivity of the agro-environmental measures in relation to the conservation of plant diversity in the Mediterranean region has been limited by the effects of the landscape complexity and the land-use management intensity at different spatial scales (Concepción et al. 2008).

The conservation of segetal species is controversial because their preferred habitat, cereal fields, are subject to productive and economic interests, which leads to the permanent conflict between crop production and diversity conservation (Fried et al. 2009; Foley et al. 2011). However, it is necessary to allocate efforts to the conservation of biological diversity in agricultural systems. For this reason it is important to evaluate the status of the segetal species populations that are rare in the Mediterranean region and understand their response to the farming practices to establish the appropriate measures of conservation.

3. Study design

This thesis pretends to improve the current knowledge of the characteristic arable species of dry land cereal fields (communities of the phytosociological order *Secalietalia cerealis* Br.-Bl. 1936) and to evaluate why some of these species have become rare in these habitats. To this end, the study has been structured around two main axes that pretend 1) to assess the frequency of characteristic arable species, and especially of those that have become rare, in relation to the management of dry land cereal fields of Catalonia (*Chapters 1* and *2*) and 2) to evaluate the effects that some particular agricultural practices have on the fitness of certain segetal species (*Chapters 3* and *4*).

3.1. <u>Analysis of the status of characteristic and rare arable species in the dry land cereal fields of Catalonia</u>

3.1.1. Study area: dry land cereal arable areas of Catalonia

The study on the status of segetal species' populations was carried out in 2011 in the dry land cereal cropping area of Catalonia, mainly in the Central Depression. The study area spans approximately 100 km × 80 km (from 41º 22' to 42º 06' N and from 0º 59' to 2º 12' E), and with an average altitude (± SE) of 558 ± 30 m a.s.l. (Figure 4). The climate of the area is continental Mediterranean, with mean annual temperatures of 12.6 ± 0.2ºC (between 10.5 and 14.9ºC) and average annual precipitation of 637 ± 21 mm (from 416 to 868 mm) (Ninyerola et al. 2005). The variability in rainfall and temperature parallels a climatic gradient in the study area, with dryer conditions towards the western sector. Generally, the whole area lies on marl and calcareous sandstone, on which basic soils develop.

The agricultural landscape of the study area is a complex mosaic of agricultural areas with fragments of semi-natural and natural habitats, and also urban and industrial areas and roads. The natural habitats in this area include pine (*Pinus halepensis* Mill. and *P. nigra* Arnold subsp. s*alzmannii* (Dunal) Franco) and oak (*Quercus ilex* L. and *Q. faginea* Lam.) woodlands as well as scrub, small stands of perennial-dominated grasslands and riverine vegetation. At the field boundaries, however, it is more common to find bramble thickets, dry grasslands (dominated by *Brachypodium retusum* (Pers.) P. Beauv. or *B.*

phoenicoides (L.) Roem. et Schultes) and various kinds of ruderal vegetation. Agriculture is the main land use in this area, dominated by dry land cereal crops mostly sown with barley (*Hordeum vulgare* L.) and wheat (*Triticum aestivum* L.), and to a lesser extent with oat (*Avena sativa* L.) and spelt (*Triticum aestivum* L. subsp. *spelta* (L.) Thell.). These crops constitute the agricultural mosaic together with forage crops, especially alfalfa (*Medicago sativa* L.) and other annual rain-fed crops such as rapeseed (*Brassica rapa* L. subsp. *oleifera* (DC.) Metzg.) and sunflower (*Helianthus annus* L.), but also vineyards (*Vitis vinifera* L.) and almond (*Prunus dulcis* L.) and olive (*Olea europaea* L. var. *europaea*) fields, among others (Departament d'Agricultura Alimentació i Acció Rural. Generalitat de Catalunya 2009).

The plant communities in the annual crops considered in this study are traditionally included in the phytosociological order *Secalietalia cerealis* Br.-Bl., 1936. However, in the study area there are mostly those *Secalietalia cerealis* communities developing on basic soils. These communities are dominated by annual species such as the poppy (*Papaver rhoeas* L.), field gromwell (*Lithospermum arvense* L. subsp. *arvense* L. = *Buglossoides arvensis* (L.) I. M. Johnst. subsp. *arvensis*), lady's Venus comb (*Scandix pecten-veneris* L. subsp. *pecten-veneris* L.), yellow crownvetch (*Coronilla scorpioides* (L.) Koch), hound's ear (*Bupleurum rotundifolium* L.) or hedgehog parsley (*Caucalis platycarpos* L.), among others.

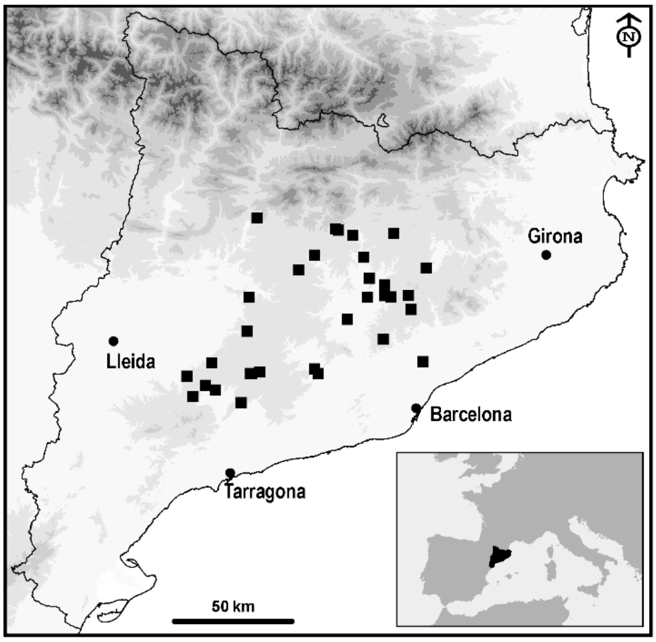

Figure 4: Location of the study sites in Catalonia. Squares indicate the farms surveyed. Grey tones indicate the elevation every 500 m.

3.1.2. Experimental design. Chapters 1 and 2

We selected a total of 304 fields from 32 farms (Figure 4). We only considered organically managed farms to increase the probability of detection of the segetal species (Romero et al. 2008b; José-María et al. 2010). Some organically managed farms with dry land cereal crops were selected outside the Central Catalan Depression. Farms were distributed in a gradient of landscape complexity, assessed as the mean percentage of arable land (PAL) in areas of 1 km radius around each field of each farm. Landscape structure of these farms ranges from structurally simple, with high values of PAL (around 97 % of arable land), to complex landscapes with predominance of natural and semi-natural habitats (only 8 % of arable land) and, thus, where agricultural pressure is lower (Figure 5).

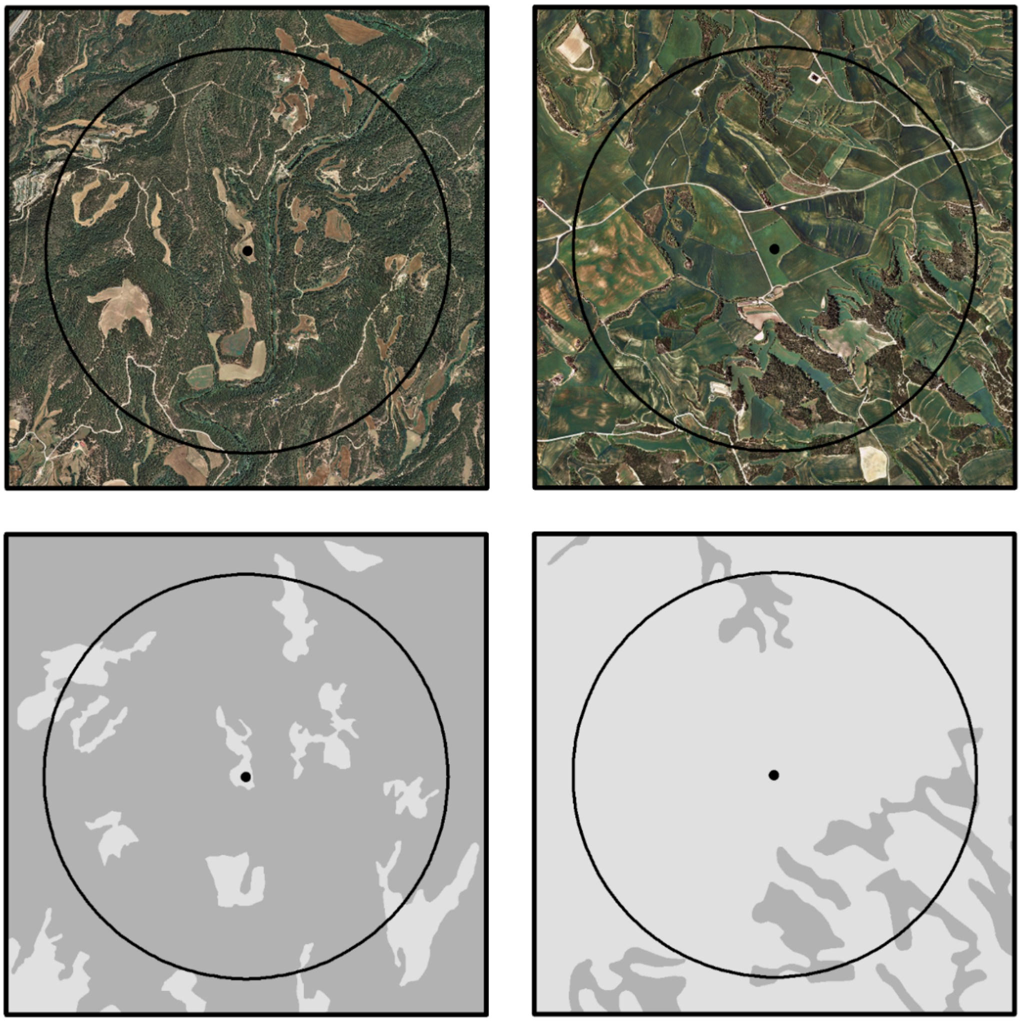

Figure 5: Examples of sampled fields (black dot) representative of a complex landscape (left) and a simple landscape (right). The upper panel shows the orthophotomaps of the surroundings of the fields and the areas of 1 km radius around each one. The lower panel shows the Catalan Habitats Cartography converted to a binary map, with agricultural habitats in light grey and the natural and semi-natural habitats in dark grey.

The percentage of arable land (PAL) is a widely used measure of agricultural intensification at a landscape scale. The PAL correlates with other landscape descriptors such as the habitat diversity and fragmentation, the density of boundaries or the extension of semi-natural habitats (Roschewitz et al. 2005). Furthermore, these correlations have been found in contrasted landscapes from the north and centre Europe and from the Mediterranean, for instance (Roschewitz et al. 2005; Gabriel et al. 2005; Romero et al. 2008a).

Within each farm, we selected the fields that had been sown, during the season previous to the sampling, with the annual crops usually included in the crop rotation of winter cereals: cereals for grain or forage, legumes also for grain or forage and polycultures containing cereals and legumes. Multiannual crops were avoided because of their heterogeneity at the sowing time. Between four and twelve fields per farm were surveyed, depending on the number of fields that met the criteria of selection. With the aim to maximize the detection of characteristic arable species and of the ones being rare, the sampling was restricted to the field edges. We evaluated the presence of the segetal flora within sampling plots established 80 m apart along the field edge (Figure 6). In that way, we surveyed a minimum of three and a maximum of ten plots per field depending on the

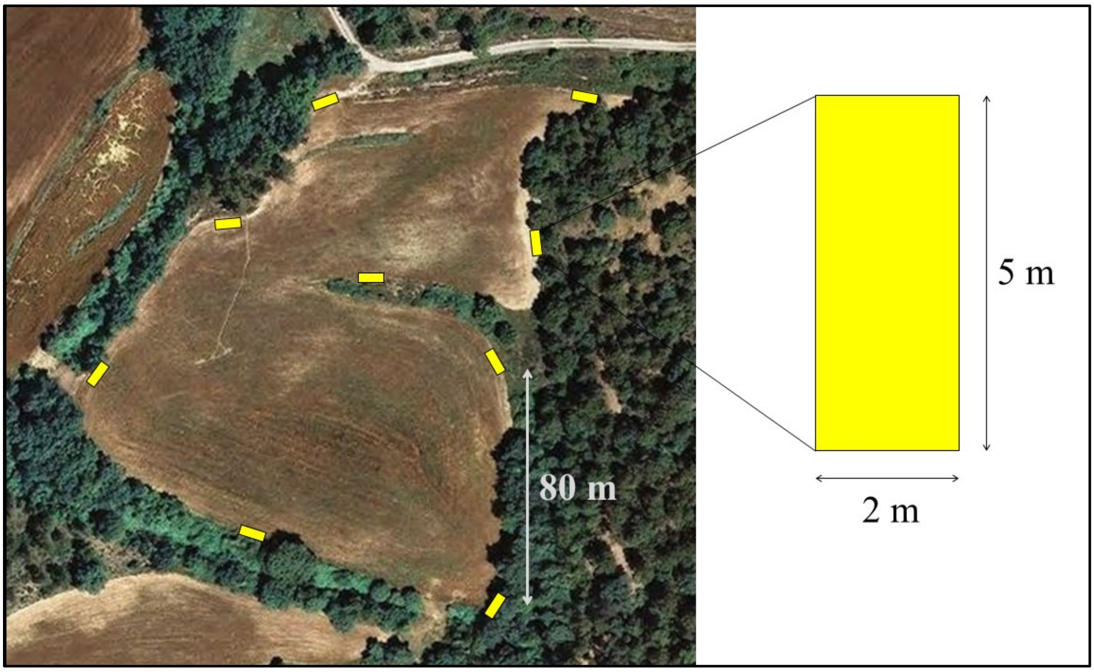

Figure 6: Scheme of the distribution of sampling points in a field. The sampling points were located at the edges of the field, separated 80 m apart. At each sampling point the presence of characteristic species was evaluated in an area measuring 2 m × 5 m.

field size. Within each plot, we recorded the segetal species appearing in an area of 2 × 5 m parallel to the margin. We also recorded different characteristics of the sampled plot such as the crop and the total weed cover and the habitat type of the adjacent margin categorized into woody vegetation, grasslands, ruderal vegetation and others.

The information on field management was obtained by means of personal interviews to the farmers. These interviews allowed us to acquire the information about their farming practices during the last season, specifically the type of tillage, the crop sown, the seed origin, the sowing time and density and whether the field was grazed or not. We also collected information about the years since the conversion to organic farming and about the crops sown during the last five years. The entire management of the farm was also considered, in terms of farm production orientation (crop production specialist or farms with crops and livestock) and the proportion of cereal crop fields per farm in the current year.

Landscape structure was also characterized through the percentage of arable land (PAL) within circular areas of 1 km radius around each field. The field physical characteristics were also defined with measures such as the area and the shape (area/perimeter) of the fields. These measures were taken from colour orthophotomaps and from the Catalan Habitats Cartography (Carreras & Diego 2004) using ArcGIS (version 9.2). Landscape structure of the farms was also considered through the average PAL around the fields of the farm and through the mean distance between fields of the farm.

These data allowed the assessment of the effects of the different organic farming practices at the field and farm levels on the characteristic arable species diversity and on the subset of the segetal species considered rare, taking into account their correlations with the landscape complexity and the field area and shape as well (*Chapter 1*). This diversity was analysed, in *Chapter 1*, using the multiplicative partition of diversity (Allan 1975), which allows the partition of the overall diversity into independent components (Jost 2007). Therefore, the species richness at the farm level (γ diversity) is broken down into the mean contribution of each field (α diversity) and the diversity owing to the divergence among community compositions of the fields (β diversity, calculated as $\beta = \gamma / \alpha$). We also carried out a more detailed analysis assessing the effects of farming practices, landscape and field physical descriptors on the probability of finding these rare segetal species in a certain field, as well as the plot characteristics and the habitat of the adjacent margin that favour the presence of rare arable species (*Chapter 2*). These analyses were also conducted for each one of the most frequent rare arable species to disentangle how these species respond individually to the variables considered and whether there are common patterns of response (*Chapter 2*).

3.2. Evaluation of the effects of specific farming practices on the fitness of some characteristic arable species

3.2.1. Experimental design. Chapters 3 and 4

We conducted two experiments under controlled conditions to assess the effects of specific farming practices on the growth and reproduction of characteristic arable species. With these studies we have evaluated the impact of herbicide application (*Chapter 3*) and of fertilization (*Chapter 4*) on different pairs of characteristic arable species belonging to different families. The species selection took into account that the species within each family differed on their frequency in the study area, being one rarer and the other more common. This test allowed us to take into account the fact that close relatives may respond similarly to particular environmental conditions, which makes fairer the comparison between common and rare species.

In *Chapter 3*, we analysed the effects of the herbicides 2,4-D and tribenuron applied at six different doses plus controls on four pairs of characteristic arable species, each pair including one rare and one more common species from a different family (Apiaceae: *Bupleurum rotundifolium* L. and *Scandix pecten veneris* L. subsp. *pecten-veneris*; Brassicaceae: *Neslia paniculata* (L.) Desv. subsp. *thracica* (Velen.) Bornm and *Rapistrum rugosum* (L.) All.; Papaveraceae: *Papaver argemone* L. and *P. rhoeas* L.; Rubiaceae: *Asperula arvensis* L. and *Galium aparine* L. subsp. *spurium* (L.) Simonk). With this experiment we assess the effects of herbicides on the growth and on the ability to produce seeds of the arable species tested at a short term endpoint (28 days after the herbicide application) and at a long term endpoint (48 or 55 days after the application). We also evaluated the ability of these species to recover following sublethal herbicide exposures. It was also assessed whether the differences in the responses between the species in the same family correlate with the differences in their frequency in the region.

Given that the fertilization has important effects on the growth and reproduction of characteristic arable species through the competitive interactions between crop and arable weeds, the test to characterize the effects of fertilization was assessed together with the effects of competition with the cereal. In the *Chapter 4* we evaluated the effect of the type (organic and mineral) and the dose (high and low) of fertilizer application on four pairs of common and rare characteristic arable species paired according to the family to which they belong (the same pairs of species as in *Chapter 3* but the Apiaceae; *Bulpeurum rotundifolium* L. was replaced by *Bifora testiculata* (L.) Spreng.), when growing with or without wheat (Figure 7).

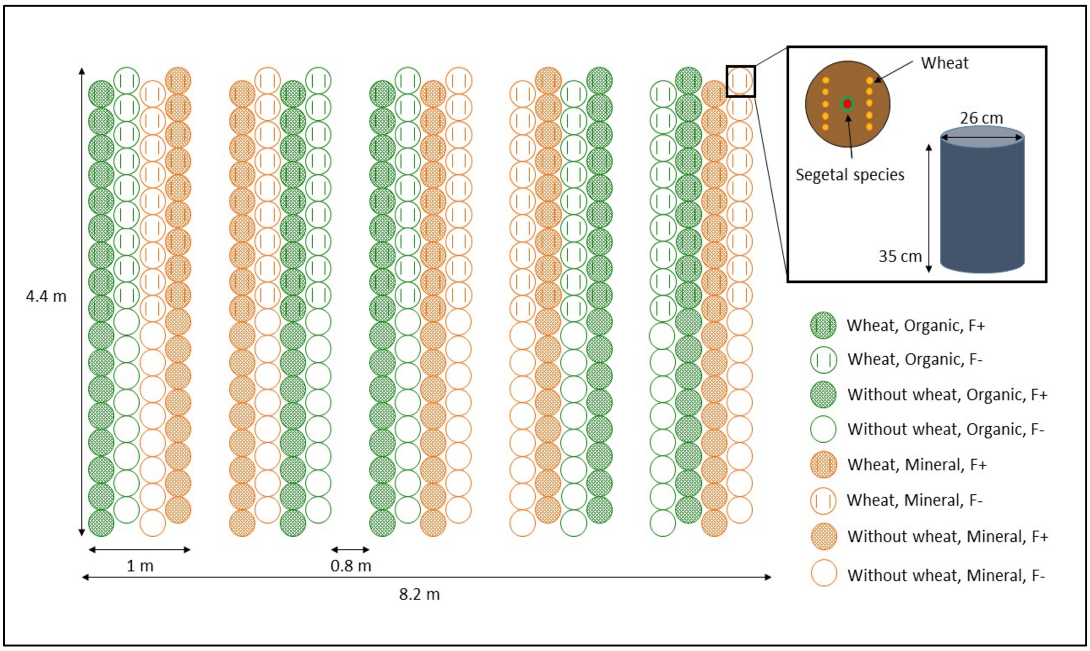

Figure 7: Scheme of the experimental design used for testing the effects of the type (organic and mineral) and the dose (high, F+ and low, F-) of fertilization and crop competition (wheat and without wheat) (*Chapter 4*) depicting the distribution of the pots according to the treatment. On the top right, a pot is represented in the top and lateral view, displaying the dimensions and the distribution of wheat and segetal species' plants in each pot.

Objectives

1. General aims

The main objective of this PhD thesis is to study the status of the arable species populations that are characteristic of Mediterranean dry land cereal fields in relation to the farm management and to the landscape structure and assess the reasons why some of them may have become rare in these agricultural habitats. To this end, the study has been divided into two main complementary research lines, one observational and another one experimental, with the aim to:

- Determine the effects of farming practices and the contribution of the landscape structure on the characteristic arable species, and particularly of those that are considered rare, on dry land cereal crops. To this aim, we evaluated the presence and the species richness of characteristic arable species at the edges of organically managed fields in the dry land cereal area of Catalonia.

- Evaluate the response of some characteristic arable species to specific farming practices such as herbicide application and fertilization together with cereal competition. At the same time, we pretended to check how the rarity degree of these species depends on their fitness in response to these farming practices.

2. Specific aims

The specific objectives of each chapter of the current thesis are detailed below:

Chapter 1:

i) To assess the effects of both current and past (in the last five years) farming practices, field size and shape, and landscape structure on the species richness of characteristic and rare arable weed species (α diversity) in organically managed fields.

ii) To evaluate the effects of management at the farm level, defined as the set of practices affecting the farm as a whole, apart from the surrounding landscape variables, on the farm diversity (β- and γ-diversity) of these sets of species.

Chapter 2:

i) To assess the effects of farming practices, field and landscape structure and local conditions on the occurrence of rare segetal species at the edges of the organically managed fields of the Central Depression of Catalonia.

ii) To investigate the similarities between the responses of selected rare arable species to the different variables of management and surrounding landscape assessed.

Chapter 3:

i) To examine the patterns of sensitivity of selected characteristic arable weed species to two commonly used herbicides in the Mediterranean cereal crops (tribenuron and 2,4-D) at short-term and long-term endpoints (total aboveground biomass, seed biomass and number of seeds).

ii) To determine the levels of and the time to recovery, in terms of both stem length and fruit number, of the plants affected by increasing the doses of these herbicides.

iii) To assess whether the sensitivity to these herbicides application is higher in rare characteristic arable weeds than in more common related species.

Chapter 4:

i) To ascertain the effects of the type and of the dose of fertilization on the growth of selected characteristic arable weed species and how these effects are modulated by the competitive relationship with the crop.

ii) To determine whether the flowering onset of characteristic arable species is influenced by the different fertilization rates and types and their interaction with crop competition.

iii) To assess how the competitive ability of rare arable weeds differs from the more common species that are phylogenetically related, at the different fertilization types and doses.

General discussion

Several studies have pointed out that the agricultural intensification at both the local and the landscape scales has notably affected the characteristic arable, or segetal, species populations of dry land cereal fields. The intensification has caused quite significant population declines in many species, which nowadays are mainly found in the field edges (Romero et al. 2008b; José-María et al. 2010). However, it has often been highlighted the lack of specific knowledge of the effects that agricultural intensification may have on characteristic arable species due to its low frequency and abundance. With this study we partially overcame this limitation by surveying the characteristic arable flora in a large number of plots (1957) at the edges of organically managed fields (304), which allowed us to maximize the detection of characteristic arable species. This sampling did not cover the whole spectrum of agricultural management given that it has been previously shown that the presence of most of these species is extremely low under conventional farming (Romero et al 2008b; José-María et al. 2010). Therefore it was an a priori decision to focus on the fields with a management that may favour their presence. This allowed us to improve our knowledge on the status of characteristic arable species populations in the dry land arable fields and relate their presence to the agricultural practices and to the landscape structure. The knowledge of the factors at the field and landscape scale, either management practices or field and farm physical characteristics, conditioning the presence of these species may help to define sustainability indicators of agricultural management aimed at biodiversity conservation, particularly for the species that have become rare (*Chapters 1* and *2*).

The role of specific management practices on the fitness of characteristic arable species of dry land cereal fields has also been dealt experimentally. We delved into the effects that the application of herbicides and fertilizers, as examples of commonly used farming practices, may have had on the decline of characteristic arable species populations and on determining the rarity of some of them (*Chapters 3* and *4*). The knowledge acquired with these studies should enable the development of strategies to harmonize the agricultural production and the preservation of the flora characteristic of dry land cereal cropping habitats.

1. Segetal species status in the dry land cereal fields of Catalonia

Our survey of the segetal species in the Central Catalan Depression allowed us to detect more characteristic arable species than other studies on plant diversity from the same area (Romero et al. 2008b; José-María et al. 2010). Nevertheless, most of these species were found at very low frequencies. Even some species that are not considered rare by de Bolòs et al. (2005) were found at very low frequencies. With this survey we detected more than the 70 % of the rare characteristic arable species listed in the study area albeit with extremely low frequencies; many of them were found in less than 1 % of the samples. These results confirm the current scarcity of these species, as pointed out previously (Chamorro et al. 2007), while provide some hope regarding their conservation because many of them are still present in the arable fields of Catalonia.

1.1. Effects of the surrounding landscape and field physical characteristics on the segetal species

Several studies pointed out that fields embedded in more complex landscapes, usually host higher characteristic arable species diversity (Roschewitz et al. 2005; Gabriel et al. 2005; José-María et al. 2010) because the adjacent habitats might represent alternative habitats that act as a source of propagules for field recolonization. We found similar results in Catalan dry land organic arable fields for the overall characteristic arable species diversity but not for the rare segetal species subset, to which the percentage of arable land correlates positively (*Chapters 1* and *2*). The different response pattern of the rare segetal species with respect to the whole set of segetal species suggests that the neighbouring non-agricultural habitats may not benefit the rare segetal species as a source of propagules because they are more habitat specialist than the whole set of segetal species considered in this study (Meyer et al. 2015).

The landscape structure comprises characteristics at the farm scale that affect the segetal species diversity. The spatial distribution of the fields within a farm (mean distance between the fields of a farm) determines higher floristic heterogeneity (β diversity) between the fields as expected, because it may reduce the exchange of diaspores. Yet the field distribution does not affect the overall farm diversity or γ diversity (*Chapter 1*). Thus, the total characteristic arable species diversity at the farm scale is mainly determined by the conditions provided by the fields themselves rather than by the farm structure or by the surrounding landscape.

Other landscape structuring processes that affect the arable flora, such as field physical characteristics, also have a significant effect on the characteristic arable flora. Larger fields, probably originated from the historical amalgamation of smaller fields, have a richer segetal flora (*Chapter 1*), as well as higher probability of rare arable species occurrence (*Chapter 2*) than smaller fields, because merging fields increases the local set of species (Marshall 2009). Moreover, larger fields contain a broader range of microenvironments while still enabling intra-field exchange of diaspores. Instead, we had to reject the initial hypothesis according to which we expected more characteristic arable species diversity in fields with more complex shapes, with longest edges in relation to the total area, because these species thrive preferably in these areas. This result probably reflects the relevance of the area on the flora of the fields.

At a smaller scale, the effect of the vegetation at the neighbouring boundaries on the presence of segetal species at the field edges was not so important. The rare segetal species are likely to be negatively affected by habitats at the adjacent boundaries that are dominated by more competitive species, such as forests and shrublands (*Chapter 2*). The remaining habitats considered in the analysis do not seem to have a distinct effect on the segetal species. These results show that these species respond more to local in-field conditions rather than to the adjacent habitats as a source of propagules.

1.2. The segetal species and farm management

The organic management of dry land cereal fields have positive effects on the diversity of the arable species and, moreover, these effects are cumulative over time. For this reason, the fields and farms that have been managed organically for a longer period host higher diversity of characteristic arable species (*Chapter 1*). However, the intensity of organic farming practices is highly variable (Clough et al. 2007; Armengot et al. 2011), which leads to a large versatility of the overall effects of organic farming on diversity. For this reason, it is essential to know the effects of particular farming practices on the characteristic arable species. Most farming practices operate, primarily, at the field scale, but other practices operate simultaneously and in the same way over all the fields of a farm. This fact may have consequences on the overall diversity and on the heterogeneity of the floras between fields in a farm (*Chapter 1*).

Characteristic arable weeds are mainly annual species whose persistence depends on the soil seed bank and on the ability to germinate, establish and replenish again the soil seed bank. They are predominantly autumn germinating species that take advantage of the crop sowing, typically cereals, and complete their life cycle during the crop development. The remarkable coupling of the biological cycles of the segetal species with the seasonality of agricultural management of winter cereal cropping systems highlights the importance of the crop type for the maintenance of segetal species populations. Thus, the standing crop and the associated crop management represent important filters on the presence of characteristic arable species (Marshall 2009; Fried et al. 2009).

Disturbance regime of agricultural habitats is critical for segetal species establishment, which are both affected by the intensity and the nature of disturbance (Critchley et al. 2006). Tillage represents the major disturbance in arable systems. Typically, the tillage in these systems is carried out at the end of summer or during the autumn, before the crop sowing. This disturbance regime matches the typical germination requirements of most of these characteristic species of dry land cereal crops (Saatkamp et al. 2011). For this reason, autumn sowing and particularly the sowing of cereal crops provides the conditions favouring the presence of characteristic arable species and of the rare ones (*Chapters 1* and *2*). Tillage intensity significantly affects the segetal species establishment as well. When soil tillage is performed without modifying the vertical soil structure (i.e. chisel or cultivator) it encourages the presence of characteristic arable species and of the rare species because it offers favourable conditions for the germination of the seeds from the upper soil layers and of the ones arriving from dispersion. Instead, the soil-inverting tillage (as the mouldboard plough) promotes deep seed burial, which prevents the germination and emergence and, therefore, the establishment of new individuals (Gruber & Claupein 2009). Other sources of disturbance, like stubble grazing, have a negative effect on the segetal species diversity (*Chapter 1*). Although these species are adapted to soil disturbance, they cannot withstand the disturbance caused by herbivory.

Therefore, cereal crops promote higher characteristic arable species diversity when they are sown (*Chapter 1*), at the same time that benefit the occurrence of rare arable species

(*Chapter 1* and *2*). This is a sign of the adaptation of the segetal species to cereal crops (Kolářová et al. 2013). As most of the arable species that germinate and thrive in a particular year come from the soil seed bank, rotations including a high proportion of cereal crops allow higher entrance of seeds of characteristic arable species to the soil, which encourage the maintenance of these species' populations. Since cereal crop fields hold more characteristic arable species, farms having more fields simultaneously sown with cereal crops would sustain higher overall richness of characteristic arable species than farms with more diversified crops, in general. Thus, the fields are richer although they hold a more homogeneous flora (the β diversity is lower in farms with higher proportion of cereal crops) probably due to the more frequent exchange of seeds by the agricultural machinery (Armengot et al. 2012). The heterogeneity of the flora between the fields of a farm increases in relation to the management diversity among fields, which happens frequently in farms also keeping livestock. Nevertheless, this floristic heterogeneity among fields of a farm does not compensate the decrease of the overall farm species richness because of the lower diversity of the fields. For this reason, efforts should focus in promoting the diversity of segetal species and of the rare species at the field level.

Beyond the establishment, the persistence of segetal species depends on their ability to thrive in an environment dominated by crop plants. Arable species and crops compete for the same resources and thus they tend to be mutually exclusive (González Ponce 1998). But in general, characteristic arable species of dry land cereal habitats tend to be poor competitors within crops (Critchley et al. 2006). Thus, the farming practices encouraging the crop growth affect them negatively. Field fertilization represents an important determinant of the competitive relationships between the crop and the arable species (Goldberg & Miller 1990; Iqbal & Wright 1997; Tang et al. 2014). High applications of fertilizers increase the nutrient availability for plants. Nevertheless, crop plants usually uptake the nutrients and grow faster than the segetal species. Therefore, crop plants efficiently occupy the space during the establishment and reduce light penetration, which reduces the possibility of growth and development of the characteristic and rare arable species (*Chapter 4*). This negative effect of the high fertilizer applications is common on segetal and rare species. Therefore, these species are less frequent and their diversity is lower in the dry land cereal fields with higher organic fertilizer inputs (*Chapters 1* and *2*).

However, organic fertilization, which releases the nutrients slower than the mineral fertilization, generally favours the development of characteristic arable species and of the rare species. The negative effects of the crop competition are lowered under these conditions, because the cereal crop grows less vigorously than under mineral fertilization (*Chapter 4*). Therefore, the application of appropriate amounts of organic fertilizers would be a good strategy to promote the segetal species conservation while, although reduced, it does not threat the crop yield.

The edges of the fields are characterised by lower fertilizer inputs and by more sowing failures and poorer establishment of crop seeds. Therefore, the crop cover there is generally low. When the local conditions are favourable, they allow a good development of both the crop and the characteristic and rare arable species (*Chapters 1* and *2*). This means that, under the conditions in which the sampling was performed, there is a positive correlation between the presence of these species and the crop cover. This correlation between crop and weeds is also manifested in the variability within the fields; despite the field is the main unit of management, inhomogeneity of the farming operations across the field involves certain variability in the crop cover (related to the sowing density and the establishment) and the total weed cover. Therefore, at the very local level, where the crop establishes and thrives properly owing to somewhat favourable local conditions, the rare segetal species are benefited as well (*Chapter 2*), contrary to the expectations.

2. Response of the segetal species to herbicides application and fertilization

Many characteristic arable species are largely absent from the conventionally managed fields because of the intensive farming practices that characterise these systems (Romero et al. 2008b; José-María et al. 2010). Herbicide application has been considered one of the main causes of the decrease of plant species diversity in agricultural systems (Hyvönen & Salonen 2002; José-María et al. 2011). Herbicides can even affect the vegetation of the adjacent habitats, which are reached by application drift (Riemens et al. 2008; Dalton & Boutin 2010; Bassa et al. 2011; Schmitz et al. 2014). The high specificity of the segetal species to the dry land cereal habitats and the continued pressure from the application of herbicides in the conventionally managed farming systems may have negatively affected their populations. Besides, continued application of high amounts of fertilizers may have also represented a key factor determining the species diversity by altering the competitive relationship between the crop and the arable species (Goldberg & Miller 1990; Iqbal & Wright 1997; Tang et al. 2014).

Tribenuron and 2,4-D, two commonly used herbicides in dry land cereal fields of Catalonia, have important effects on the biomass and reproduction of arable species, even at low doses of application (*Chapter 3*). The effects of herbicides application depend on the dose of application and on the type of herbicide applied (Clark et al. 2004; Carpenter & Boutin 2010) but there is considerable variability in the response between species. Some segetal species, however, are able of a certain recovery depending on the herbicide and on the dose of application, as pointed out previously (Riemens et al. 2009; Carpenter et al. 2013). Therefore, those more affected species or with low ability to recover would be displaced at the expense of those more tolerant, resistant or faster at recovery (Carpenter & Boutin 2010; Carpenter et al. 2013; Egan et al. 2014), or would simply be competitively excluded by the crop (Christensen 1994; Williams et al. 2008). Hence, the effects of herbicides must be considered within a community context.

Higher mineral fertilizer inputs do not represent a limitation on the growth of the characteristic arable species when growing alone. This fact indicates that the negative effect of fertilization on the segetal species operates through the competition with the crop (*Chapter 4*). In this way, the cereal displaces the segetal species by means of faster resource uptake and more efficient use (Robinson & Sutherland 2002). Contrarily, organic fertilization, which releases slowly the nutrients, reduces the negative effects of the crop on the characteristic arable species and on the ones that are rare.

Given that most segetal species are annual species, farming practices that affect their reproduction decisively influence the ability of characteristic arable species to thrive in a particular year. Both the herbicides application and the fertilization may involve noticeably negative effects on the flowering or on the fruit set, which may reduce the seed production with the subsequent impacts on the seed bank replenishment (Strandberg et al. 2012; Qian et al. 2014). The characteristic arable species subjected to sublethal herbicide doses generally suffer more noticeable reductions in the seed production than on the total biomass, as previously described (Clark et al. 2004; Carpenter & Boutin 2010; Boutin et al. 2014; EFSA Panel on Plant Protection Products and their Residues 2014).

However, the flowering onset of the characteristic arable species did not show a consistent pattern of response in relation to the competition with the cereal crops under the different fertilization regimes. Both the low fertilization and the organic fertilizer inputs tend to cause delays on the beginning of flowering, but rarely this effect is statistically significant. However, this might have important implications on the species' populations because when species bloom later, they tend to produce fewer offspring (Storkey 2006; Fried et al. 2012).

The results of these studies under controlled conditions are of great importance because they allow us to evaluate the individual effects of specific practices on the development of certain species of interest. At the same time, they avoid the interference from other variables that can hinder the understanding of the results (Dalton & Boutin 2010). In addition, they are absolutely irreplaceable to quantify the effects of certain farming practices (the application of herbicides and fertilizers) on the segetal species and, particularly to disentangle the effects occurring under conventional agriculture, the most widespread farm management system. The low frequencies of the characteristic arable species currently found within the conventionally managed fields, on one hand, hinder the detection of these species and, on the other hand, make impossible a precise estimation of the effects. However, the results of controlled experiments should be taken with caution, especially when extrapolating the results to the natural communities functioning. The coexistence with other species, the lack of transitivity in competitive relationships and the temporal and spatial variability, among others, are the logical limitations to which any experimentation is subjected.

3. Determinant factors of rarity of segetal species

Herbicide application has been considered one of the main causes of the decrease of arable species diversity in agricultural landscapes (Hyvönen & Salonen 2002; José-María et al. 2011). Therefore, we expected that those species currently appearing under low frequencies in the fields may be more sensitive to the herbicides than those more common species of the same family. Similarly, as the high fertilizer inputs enhance the growth of the cereal crops at the expense of those species of less competitive ability, we assumed that the rare species would be disadvantaged under these conditions (Storkey et al. 2010;

Kovács-Hostyánszki et al. 2011). However, contrary to our initial hypothesis, the experimental results are not conclusive on the relationship between the rarity and the sensitivity to herbicides or the competitive ability with the crop at different fertilizer doses and types (*Chapters 3* and *4*). For some pairs of species tested, the rarer species was effectively more sensitive to a certain herbicide or more affected by the competition with the crop at higher doses of mineral fertilization than the common paired species. But in other cases, the more common species resulted more affected or both species of the same family responded similarly.

Both the application of herbicides and fertilization at high doses have been important factors involved in the general loss of arable species diversity. Therefore, it is advisable to reduce the use of herbicides and of fertilizers, particularly the mineral ones, at the edges of the fields managed conventionally to facilitate the conservation of the segetal species. However, the sensitivity to herbicides and the competitive response under different fertilization regimes are poor predictors of the rarity of the characteristic arable species individually. The extremely low frequencies of some segetal species are likely to be caused by several factors, isolated or in combination, that have affected the various arable species differently.

The assessment of the presence of the rare segetal species in the dry land cereal fields of central Catalonia allows us to reconsider what is the extent of the relation between the agricultural practices, landscape structure and the rarity of some of these arable species. It is difficult to establish solid statistical models discerning which conditions and farming practices significantly affect the presence of individual rare segetal species because, even when selecting the most frequent ones, they are still unusual species. Nevertheless, characteristic arable species in general show common patterns of response to the different variables considered in the model (*Chapter 2*). Conditions that correlate negatively with the diversity of characteristic arable species also negatively affect the probability to find the rare segetal species, as it has been previously reported (Romero et al. 2008b). This common trend for the rare characteristic arable species suggests that in general they tend to appear preferentially under particular specific farming practices. Hence, similarly to the overall segetal species diversity, sowing cereals, especially of autumn-sowing varieties, after no-inversion soil tillage may favour the conservation of these species on organically managed fields.

However, the patterns that determine the presence of the rare characteristic arable species are less obvious than the patterns determining the species richness. The random factors both at the field and at the farm level are often more important on determining the rare arable species occurrence than the local conditions or the management and landscape variables considered (*Chapter 2*). Therefore, in addition to these variables, the presence of these species depends, to a great extent, on the field and farm themselves. That is, the presence of the rare segetal species seems to be the result of the interaction of stochastic and historical factors of the fields and of the farms, that determine the set of local species (Hiltbrunner et al. 2008), with the management, which conditions their expression in a certain year. Therefore, the segetal species conservation actions should focus on those areas where these species have been already found to avoid their loss.

4. Implications for conservation

The segetal species are undoubtedly one of the groups of species that has experienced a sharper decline in Catalonia (Chamorro et al. 2007) and that, by now, raise most conservation issues in many European countries (Sáez et al. 2011). The repeated annual applications of different herbicides to the dry land arable fields to control the weed species has been one of the main causes of the decline of arable plant diversity because they have important effects both on their growth and on their reproduction. Moreover, fertilization has also impacted the presence of these species by favouring the crop growth (and probably also the more competitive weed species) at the expense of the characteristic arable species. However, the particularly low frequency of rare segetal species is likely to be the consequence of several factors, and probably the consequence of the interaction of some of them, whose effects may vary from one species to another. Therefore, the promotion of management measures aimed at the conservation of the rarer arable species requires specific studies that allow discerning which farming practices are the most influential in each case.

The organic management seems an appropriate measure to favour the characteristic arable species, which are still present at the edges of the fields. However, these species actually occur under very low frequencies, even within the organically managed fields in the cereal producing region of the central Catalonia. For this reason, we consider that it is the right moment to implement measures for their conservation. The foundations for the conservation of characteristic arable species should go beyond the organic management, because some organic farming practices do not benefit the segetal species. Specific farming practices such as the sowing of cereal crops, especially in autumn, after no-inversion soil tillage should be encouraged to promote the characteristic arable species populations. At the same time, fertilization should be rationalized even in the form of organic fertilizers, to avoid compromising the crop production while ensuring the persistence of these arable species. Furthermore, grazing should be restricted in those areas where segetal species populations have been found, especially if they are considered rare.

Other actions aimed at the farm or landscape levels are less feasible and less likely to be effective. On the one hand, factors acting at the local scale have larger effects on the presence of characteristic arable species; on the other hand, conditions at the farm and landscape scales are less likely to be modified. Nevertheless, cereal production at the farm level, particularly in farms managing distant fields, should be promoted to develop effective conservation strategies, favouring the presence of the segetal species as well as the exchange of their diaspores and their establishment in new fields.

General conclusions

In this thesis, it has been determined the frequency and the diversity of the arable species characteristic of the dry land cereal habitats, or segetal species, at the edges of organically managed fields from the central Catalonia. It has been evaluated how these species depend on the surrounding landscape characteristics, on the field management and on the local conditions within the fields. We have determined the extent of these effects on the segetal species found in the study area, as well as on the subset of the rarest species, which require attention regarding their conservation. Moreover, it has been analysed the role of specific widespread farming practices, such as the herbicide use and the fertilization, on determining the differences in the frequency of some segetal species. These studies allowed us to know which agricultural practices should be encouraged to promote the conservation of these species.

We outline below the main conclusions from the different studies included in this thesis.

- Characteristic arable species or segetal species that have been cited in Catalonia occur at very low frequencies, even in the organically managed fields. This fact highlights their delicate status of conservation and the need to promote their conservation at this moment when most of them are still present in these fields.

- Generally, although the continuity of organic management favours the segetal species diversity, the effects of the different farming practices within organic management are quite variable. Therefore, it is necessary to evaluate individually the farming practices that might affect the characteristic arable species populations to develop the conservation strategies.

- Farming practices at the field level represent important determinants of the segetal species richness and of the rare ones. Sowing cereal crops, preferentially in autumn, after no-inversion tillage favours the characteristic arable species and, at the same time, these practices confer favourable conditions for the occurrence of most of the rare segetal species.

- Fertilization was also a factor that significantly affects the frequency of the segetal species and the rare arable species within the dry land cereal fields of Catalonia. Excessive nutrient availability stimulates crop competition at the expense of both common and rare segetal species growth.

- The percentage of arable land around each field has a negative effect on the diversity of segetal species within a field (α diversity), but positive on the presence of the rare arable species. These results highlight the greater preference of the rare segetal species for the dry land cereal habitats than the whole set of the segetal species considered.
- The field area has a significant positive effect on both the overall segetal species diversity and on the diversity of the rare segetal species, so that larger fields support more segetal species richness while there is more probability to find rare arable species. However, fields with higher perimeter/area ratio hold less segetal species.
- Overall segetal species richness of the farms (γ diversity) depends primarily on the field species richness (α diversity) rather than on the heterogeneity between the floras of the fields (β diversity). Therefore, the higher proportion of cereal crop fields, which sustain more characteristic arable species (α diversity) but homogeneously distributed among fields, defines higher farm segetal species diversity (γ diversity).
- Despite the overall farm diversity does not increase, farms with livestock have a more heterogeneous flora between fields due to the diversity in the management of the different fields. More heterogeneous flora is also found when fields are more distant between them.
- Although it is difficult to establish statistically significant models for the presence of the rare segetal species, there are similar patterns of response between the rare arable species occurrence and the landscape structure, the farm management and the local conditions evaluated.
- The rare characteristic arable species appear more frequently where the overall weed cover is higher, which indicates that the conditions benefiting the abundance of weeds also favour the presence of the rare arable species. The rare segetal species tend to avoid the crop edges next to more competitive margins such as forests or shrublands, but have no marked preference for any other habitat on the field margins, which may not act as refuges.
- The rare segetal species are more likely to appear at the field edges where the crop cover is higher, because it is usually not very high at the edges of the fields and when

the appropriate conditions for the crop occur they also benefit the segetal species growth.

- Stochastic factors that depend on the particular history of the fields and of the farms have a large effect on determining the presence of rare arable species in the fields, frequently larger than the effects of farming practices.
- The application of herbicides affects the characteristic arable species tested even at low doses of application. However, their herbicide sensitivity depends on the type of herbicide, being the responses species specific.
- In general, the short term effects of herbicides on the biomass are larger than the effects detected at the long term. This indicates a certain ability of characteristic arable weed species to recover. The recovery patterns largely depend on the dose of application, requiring more time as the doses of herbicide application increase.
- The effects of herbicides are generally more pronounced on the plant reproductive ability than on the total biomass in the long term assessment. Given that these species are annual plants that rely on reproduction to ensure future population recruitment, the short term assessment using the biomass as the only measure of the herbicide effects is not an appropriate criterion to evaluate these effects on the segetal species populations.
- Differences in the sensitivity to herbicides between phylogenetically close characteristic arable species do not explain the overall differences in the current frequency between these species.
- The segetal species growth is not hindered by high mineral fertilization, but this fertilization intensifies the negative effects of the cereal competition on these segetal species in comparison to organic fertilization. Therefore, organic fertilization may promote the segetal species development while does not undertake much the cereal production.
- Not all the rare arable species respond in the same way as compared with the common species to the cereal competition pressure under the different fertilizer types and doses.

- Despite the effects of herbicide application and fertilization have clearly contributed to the overall scarcity of the characteristic arable species, our results indicate that the differences of their effects on particular species do not correlate to their frequency in the region. The reasons of the scarcity of particular segetal species should be searched in other factors or in the combination of some of them.

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Publicacions

Capítol 1

Both farming practices and landscape characteristics determine the diversity of characteristic and rare arable weeds in organically managed fields

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RESUM

Tant les pràctiques agrícoles com les característiques del paisatge determinen la diversitat de les espècies arvenses característiques i rares en els camps de gestió ecològica

Les poblacions d'espècies arvenses característiques dels sistemes cerealistes de secà han patit una dràstica reducció, fet que ha comportat que moltes d'elles hagin esdevingut rares en els seus hàbitats preferents. Aquesta reducció ha succeït en paral·lel a una forta intensificació de les pràctiques agrícoles així com a una simplificació del paisatge. En aquest procés, les espècies segetals sovint han quedat restringides a les vores dels camps de gestió ecològica, que és on les pràctiques agrícoles hi actuen de manera menys intensa. La presència de la flora segetal és indicadora de la sostenibilitat de la gestió agrícola. Però per prevenir la seva extinció és important entendre com els diferents aspectes de la intensificació agrícola condicionen la distribució de les espècies segetals i particularment d'aquelles que són més rares.

Amb aquests motius, ens plantegem avaluar: 1) quins efectes tenen les pràctiques agrícoles actuals i passades (en els darrers cinc anys), la mida i la forma dels camps i la complexitat del paisatge sobre la riquesa d'espècies segetals i del subconjunt d'espècies rares en camps gestionats de manera ecològica (diversitat α) i 2) quins són els efectes de la gestió de les finques i del paisatge circumdant sobre la diversitat d'aquests conjunts d'espècies a escala de finca o localitat (diversitat β i γ).

Per a dur a terme aquest estudi es van mostrejar les espècies característiques dels camps cerealistes de secà (ordre fitosociològic *Secalietalia cerealis* Br-Bl. 1936), a les vores de 304 camps gestionats de manera ecològica en 32 finques (localitats) de la zona cerealista de Catalunya. Es van avaluar els efectes de la gestió de les finques i dels diferents camps, així com de les característiques del paisatge a escala de camp i de localitat, sobre la diversitat del conjunt d'espècies segetals i sobre la diversitat d'aquelles que són considerades rares. La diversitat es va avaluar en termes de la riquesa d'aquestes espècies al camp (α), a la localitat (γ) i la divergència entre les flores dels camps d'una localitat (diversitat β, calculada segons $\beta = \gamma / \alpha$). L'efecte de les diferents variables a escala de camp es va avaluar estadísticament mitjançant la inferència multi-model basada en tots els possibles models establerts *a priori*. Per a l'anàlisi estadística d'ambdues components de la diversitat a escala de localitat (γ i β), a causa del diferent nombre de mostres entre localitats, es van ajustar els models repetidament a 10.000 remostrejos aleatoris de

les dades originals. L'efecte d'una determinada variable es va avaluar a través de la distribució dels coeficients estimats.

Els efectes de la gestió agrícola i de les característiques del paisatge que es van detectar sobre la riquesa d'espècies arvenses característiques van dependre de l'escala de l'anàlisi. Els factors que operen als camps van presentar unes correlacions més fortes amb la riquesa d'espècies arvenses característiques i rares que no pas els factors que operen de la mateixa manera per tota la finca. Els anys des de la conversió a l'agricultura ecològica tenen una correlació positiva amb la riquesa d'espècies segetals. Els cultius de cereals, la sembra de tardor i les pertorbacions periòdiques del sòl determinades pel règim de llaurada afavoreixen les espècies que estan adaptades als ambients cerealistes, que tradicionalment han presentat aquestes característiques (Critchley et al. 2006; Saatkamp et al. 2011; Kolářová et al. 2013). Ara bé, la diversitat d'espècies segetals, tant en conjunt com les rares, disminueix quan la fertilització és elevada (Gabriel et al. 2005; Storkey et al. 2012), segurament pel fet que els cultius tendeixen a ser més competitius que les espècies arvenses ja que capten eficaçment els nutrients que, a la vegada, els permeten un creixement més ràpid i monopolitzar l'accés a la llum. El fet que camps més extensos mantinguin més espècies arvenses característiques i rares pot ser a causa de que ofereixen un ventall més ampli de micro-ambients alhora que permeten un intercanvi de diàspores dins del camp (Marshall 2009).

Els factors que actuen a escala de finca també van presentar una certa influència. L'augment de la proporció de camps de cereals respecte el total de camps mostrejats per localitat va correlacionar positivament amb la diversitat total (γ) i negativament amb la diferenciació de la flora segetal entre camps (diversitat β). L'estructura de la finca va tenir un efecte important sobre les diferències en la flora dels camps (diversitat β) de manera que camps més separats o gestionats de manera més heterogènia van presentar conjunts d'espècies més divergents.

Aquest estudi mostra com la gestió dels sistemes agrícoles ecològics afecta la riquesa d'espècies segetals i també d'aquelles espècies segetals que són més rares. La implementació i el manteniment de la gestió ecològica sembla una bona mesura per preservar les seves poblacions. Tot i això, a part de la conversió a l'agricultura ecològica, és necessari promoure pràctiques agrícoles específiques que reforcin la conservació d'aquestes espècies i que pal·liïn la reducció de les poblacions de les espècies arvenses més rares. L'augment de la freqüència dels cultius de cereals en els esquemes rotacionals, promoure la sembra de tardor i evitar la fertilització elevada són pràctiques recomanables per afavorir la presència d'aquestes espècies.

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Both farming practices and landscape characteristics determine the diversity of characteristic and rare arable weeds in organically managed fields

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Keywords
Crop edge; Farm management; Mediterranean dry-land arable fields; *Secalietalia cerealis*; Species richness

Nomenclature
Bolòs et al. (2005)

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Abstract

Questions: Do both current and past (short-term) farming practices and landscape characteristics have an effect on the diversity of characteristic and rare arable plant communities in organic fields? What is the role of farm management strategies, apart from farm spatial configuration, in determining the diversity components of these species sets?

Location: Thirty-two farms scattered across NE Spain (Catalonia).

Methods: Specialist species of arable fields, belonging to the *Secalietalia cerealis* Br-Bl. 1936, were surveyed at the edges of organically managed fields. We assessed the effects of farm management and landscape characteristics at the field and farm scales on α-, β- and γ-diversity values of these characteristic arable species. Analyses were also conducted on a subset of *Secalietalia* species that are considered to be rare. Statistical analyses were performed using multimodel inference determined on the basis of all possible models from an *a priori* set.

Results: Field variables, such as years since conversion to organic management, proportion of cereal crops in the rotation and autumn sowing, had a positive effect, whereas growing non-cereal crops and fertilization had a negative effect on the richness of characteristic species. The field area had a positive effect on the species richness of characteristic and rare arable plants. At the farm level, the proportion of cereal crop fields to the total amount of fields affected both β and γ characteristic diversity. The landscape variables at the farm level only influenced the β-diversity of rare species.

Conclusions: The effects of management and landscape on arable weed diversity depended on whether the field or the farm is the focus of the analysis. Characteristic and rare arable species were more affected by factors operating at local scales. Characteristic species richness responded positively to sowing cereal crops, autumn sowing and periodic soil disturbances but was negatively affected by slurry fertilization. Thus, policies promoting some of the former practices should favour characteristic arable species and mitigate the decline of the rare arable species.

Introduction

Arable weeds have been a major concern among farmers because they are traditionally viewed as an impediment to crop production, causing crop losses of approximately 30% (Oerke 2005). Thus, weed control is a primary objective guiding crop management strategies, despite the fact that only a few weed species are problematic and cause actual crop losses (Albrecht 2003). Given its current extent, agricultural land contributes significantly to global biodiversity (Tscharntke et al. 2005). Because of management intensification, however, agriculture is considered to be the primary agent for the decline of plant diversity. Agro-ecosystems are thus among the habitats that hold a large proportion of rare and endangered species in many European countries (Rich & Woodruff 1996; Tscharntke et al. 2005).

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A particular set of specialist species of arable fields belongs to the phytosociological order *Secalietalia cerealis* Br-Bl. 1936, being characteristic arable weeds that thrive almost exclusively in arable cropping systems because of their dependence on regular disturbance (Critchley et al. 2006) and their inability to succeed in more competitive habitats (Romero et al. 2008; José-María et al. 2010). In general, characteristic and rare species of any habitat are more affected by changes in land use than generalists (Albrecht 2003). Thus, the populations of characteristic arable weeds have declined severely because of agricultural intensification (Robinson & Sutherland 2002; Albrecht 2003). This decline has been so sharp that some of them have become rare or even extinct (Baessler & Klotz 2006; Chamorro et al. 2007; Cirujeda et al. 2011). Some European countries have included arable species in Red Data Lists (Cheffings & Farrell 2005; Ture & Bocuk 2008), which is a first step in developing conservation strategies (Kleijn & Van der Voort 1997). Because characteristic arable species represent key indicators of the natural and aesthetic values of farmland, their conservation in agroecosystems can be considered as an indicator of sustainable land use (Tscharntke et al. 2005; Storkey et al. 2012). The effects of agricultural intensification at field and landscape levels on the species richness of weed communities in Mediterranean dryland cereal fields have been reported extensively (Romero et al. 2008; José-María et al. 2010). Nevertheless, its effect on characteristic and rare arable weeds is still poorly understood because of their low frequency, which limits the reliability of statistical analysis. To our knowledge, this is the first study that addresses this issue by analysing the characteristic arable species present in a large sample of organic fields.

Organic farming systems, which are usually less intensively managed than conventional ones, have been found to enhance the abundance and richness of arable species (José-María et al. 2010; Ponce et al. 2011). However, large variations in management intensity among organic farms affect weed diversity (Clough et al. 2007; Armengot et al. 2011). Hence, certain current management practices might better explain the observed changes in the extant populations of weed flora than the overall intensification indicators. Because the persistence of characteristic arable weeds largely depends on their ability to remain viable in the soil seed bank, recent/past management could also help explain the diversity of weeds occurring in a given field. In addition to management, weed diversity is also negatively affected by landscape intensification (Gabriel et al. 2005; Baessler & Klotz 2006; José-María et al. 2010). Landscape intensification increases the proportion of arable habitat but also involves landscape simplification by decreasing habitat diversity and quality because of intensive field management, which is an important driver of biodiversity loss (Robinson & Sutherland 2002; Tscharntke et al. 2005).

This study aims to assess the effects of farming on characteristic weed diversity in Mediterranean dry-land organic arable fields and farms. Thus, we intend to disentangle the effects of particular management strategies that, independently of landscape characteristics, may benefit these species. We analysed characteristic arable species diversity at the field and farm (defined as the set of fields scattered in a specific area managed by the same farmer) levels. We focused on characteristic arable species, which are very specific to arable land and have suffered from recent land-use changes and may therefore have conservation value. In this way, we avoided having to include common species that also occur in neighbouring non-cropped and disturbed habitats. We surveyed organic fields from farms placed in a gradient of landscape complexity and interviewed farmers to obtain information on management practices. Specifically, we (1) assessed the effects of both current and past (in the last 5 yr) farming practices, field size and shape and landscape complexity on the species richness of characteristic and rare arable weed species in organically managed fields; and (2) evaluated the effects of farm management, apart from surrounding landscape variables, on the β- and γ-diversity values of these species sets.

Methods

Study area

Sampling was conducted in 2011 in Catalonia (41°22′–42°06′ N, 0°59′–2°12′ E) within an area spanning 100 km × 80 km (Fig. 1) with an average (±SE) altitude of 558 ± 30 m a.s.l. (min = 95 m, max = 871 m). The climate is Mediterranean, with mean annual temperatures of 12.6 ± 0.2 °C (min = 10.5 °C, max = 14.9 °C) and average annual precipitation of 637 ± 21 mm (from 416 to 868 mm). The whole area lies on marls and calcareous sandstones, which create basic soils.

All of the organically managed fields from 32 farms sown with the annual crops usually included in a crop rotation of winter cereals (cereals, legumes, ryegrass and mixtures containing cereals and legumes) were surveyed. A total of 304 fields were inventoried, with an average (±SE) of 9.50 ± 0.47 fields per farm. The farms were located in landscapes of different degrees of complexity according to their percentage of arable land (PAL); PAL is a widely used estimator of agricultural landscape simplification (Roschewitz et al. 2005; Gabriel et al. 2006). These landscapes ranged from structurally simple with a high PAL (ca. 97% within a circular area with a radius of 1 km around sampled fields; see Landscape characteristics below) to complex landscapes with a high percentage of

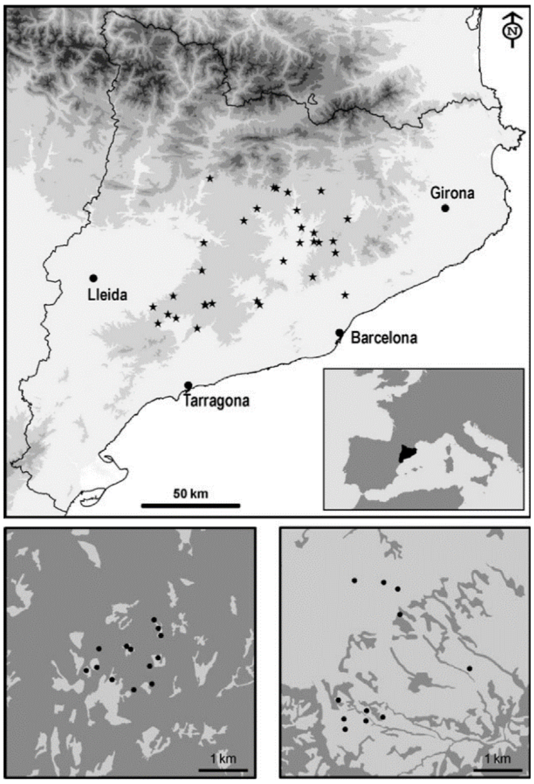

Fig. 1. Location of the 32 farms within Catalonia (NE Spain) above (grey tones indicate elevation every 500 m). Fields sampled in two farms taken as examples of complex (left) and simple (right) landscapes. Light grey represents arable habitats while dark grey includes natural and semi-natural habitats.

natural and semi-natural habitats (ca. 92%). Natural habitats in the studied area included pine (*Pinus halepensis* Mill. and *P. nigra* Arnold) and oak (*Quercus ilex* L. and *Q. faginea* Lam.) woodlands as well as scrub, small stands of perennial-dominated grasslands and riverine vegetation.

Plant species survey

The survey was conducted during May and June 2011, corresponding to the phenological optimum of weeds in the Mediterranean. The weed survey was restricted to field edges, defined as the first two cultivated meters adjacent to the margins (Marshall & Moonen 2002), to maximize the detection of characteristic arable weeds (cf. José-María et al. 2010; Kovács-Hostyánszki et al. 2011). On the edge of each field, depending on its size, a minimum of three and a maximum of ten 2 m × 5 m plots (80 m apart) were established. We used rarefaction to compensate for differences in the sampling effort between fields.

Within each plot, we only recorded arable species characteristic of the phytosociological order *Secalietalia cerealis* Br.-Bl., 1936, i.e. communities of dryland arable fields under Mediterranean climate on basic soils (see App. S1). These species were also identified as characteristic or obligate arable weeds by Albrecht (2003), Romero et al. (2008) and José-María et al. (2010). Characteristic arable weeds do not include species frequently occurring outside of the fields. Some characteristic arable species have severely declined in abundance over the last decades and are now considered regionally rare (Chamorro et al. 2007; Cirujeda et al. 2011; Solé-Senan et al. 2014). We took the subset of these characteristic species that are considered rare according to de Bolòs et al. (2005) for the analyses (see App. S1). Nevertheless, these rare arable species are not included in the Red Data List of the area surveyed (Sáez et al. 2011). The nomenclature of plant species and phytosociological adscription also follows that of de Bolòs et al. (2005). Within each plot, the percentage of crop cover was assessed.

Farming management

Farmers were interviewed to obtain information about their farming practices on each sampled field and on the entire farm during the 5 yr prior to sampling. (see Tables 1, 2, respectively). The selected variables reflect historical field management (years since the conversion to organic farming and the cereal ratio, which is the proportion of cereal crops in the 5-yr rotational schemes), and current management during the 2010–2011 period (remaining variables) of each field and farm.

Landscape characteristics

We measured the perimeter and area of each field, determined its shape (perimeter/area), and computed the PAL around it using ArcGIS 9.0 (ESRI, Redlands, CA, US). The landscape around each field was characterized within a circular area of a 1-km radius using Catalan Habitats Cartography (Carreras & Diego 2004). The mean PAL around the fields and the mean distance between the fields of each farm were considered to be the landscape metrics at the farm level.

Plant diversity

In-field characteristic and rare species diversity were evaluated as the number of *Secalietalia* species recorded in each field. The farm diversity of characteristic weeds was

Table 1. Characterization of farming practices and landscape characteristics of the fields (304) surveyed. Mean and range (in brackets) of continuous variables and proportion of fields and farms with the stated practices for the categorical variables. On the right, model-averaged estimate ± unconditional SE (UnSE) for the explanatory management and landscape variables at the field scale on characteristic and rare species richness. Asterisks indicate an effect for which the 95% CI did not include zero.

	Mean [range]/Proportion	Characteristic Arable Weeds	Rare Arable Weeds Subset
		Estimate ± UnSE	Estimate ± UnSE
Years from Conversion to Organic Farming	9.67 [1, 25]	0.078 ± 0.027*	0.012 ± 0.012
Cereal Ratio †	0.56 [0, 1]	0.272 ± 0.099*	0.155 ± 0.056*
Soil Tillage			
Inversion Tillage (vs No Till and No Inversion Tillage)	105 (199)	−0.038 ± 0.034	−0.009 ± 0.028
Current Crop			
Mixed (vs Cereal)	87 (170)	0.004 ± 0.078	0.032 ± 0.050
Legume (vs Cereal)	17 (170)	−0.161 ± 0.119	−0.056 ± 0.075
Ryegrass (vs Cereal)	30 (170)	−0.548 ± 0.110*	−0.271 ± 0.074*
Seed Origin			
Reuse Seeds (vs Purchased)	192 (112)	−0.085 ± 0.031*	−0.070 ± 0.028*
Sowing Time			
Spring Sowing (vs Autumn Sowing)	58 (246)	−0.187 ± 0.069*	−0.052 ± 0.026*
Sowing Density (kg·ha^{-1})	154.81 [16.5, 384]	0.068 ± 0.025*	0.026 ± 0.011*
Type of N Inputs			
Fertilization (vs No Fertilization)	185 (119)	−0.031 ± 0.011*	−0.017 ± 0.006*
Slurry (vs Manure)	18 (167)	−0.151 ± 0.029*	−0.097 ± 0.014*
Amount of N Inputs (kg·ha^{-1})	47.44 [0, 600]	−0.001 ± 0.006	−0.019 ± 0.010
Mean Crop Cover (%)	51.16 [0, 95]	0.056 ± 0.020*	0.099 ± 0.028*
Amount of N Inputs × Crop Cover		−0.043 ± 0.015*	−0.056 ± 0.021*
Weed Control			
Control (vs No Control)	128 (176)	0.026 ± 0.009*	0.002 ± 0.004
Tillage (Pre-sowing Control) [vs Harrowing (Post-sowing)]	89 (39)	0.110 ± 0.024*	0.043 ± 0.010*
Animal Husbandry			
Grazing (vs No Grazing)	91 (213)	−0.138 ± 0.050*	−0.068 ± 0.026*
Percentage of Arable Land (PAL)	46.58 [6.83, 100]	−0.039 ± 0.016*	0.025 ± 0.013*
Field Area (m^2)	15374.54 [1629.99, 150415.50]	0.046 ± 0.017*	0.065 ± 0.023*
Field Shape (Perimeter/Area)	0.06 [0.01, 0.19]	−0.034 ± 0.013*	−0.014 ± 0.009

†Calculated as the proportion of cereal crops in the 5-yr rotation scheme of each field.

Table 2. Characterization of farming practices and landscape characteristics of the farms (32) surveyed. Mean and range (in brackets) of continuous variables and proportion of fields and farms with the stated practices for discrete variables.

Farm Type	
Crop Specialized Farm	11/32
Mixed Farm	21/32
Ratio of Cereal Crop Fields	0.50 (0, 1)
Mean PAL	47.94 (8, 96.67)
Mean Field Distance (m)	669.87 (242.57, 3940.62)

partitioned considering the multiplicative relationship between γ species richness (total number of characteristic species in the farm) and β-diversity (accounting for the among-field community composition differentiation or within-farm heterogeneity; calculated as $\beta = \gamma/\alpha$, where α is the mean species richness per field; Whittaker 1960). The relative contributions of each fraction allowed assessment of the effects of the variables on the arable species at the farm scale.

In-field diversity analysis

As we surveyed a different number of plots in each field depending on field area, the in-field species richness was rarefied. Rarefaction provides an estimate of the expected species richness for a given number of samples. For each field, we estimated the expected number of species in three plots because this was the minimum number of plots surveyed per field.

The effects of landscape and management variables at the field scale (Table 1) on species richness were analysed using mixed effect models, which account for the hierarchically nested design. We included the farm as a random factor. Because rarefied species richness behaves nearly as a count process but its values can be fractional, we used a Gaussian model on the log-transformed values to stabilize the relationship between the variance and the mean. Due to the interaction between the amount of nitrogen input and the mean crop cover in each field detected in the preliminary analyses, this interaction was also included in the models as a new variable. This interaction variable was obtained from the product of these two variables to ensure its inclusion in the models despite the absence of main effects (Hector et al. 2010). We standardized (mean equal to 0 and SD equal to 1) all continuous explanatory variables. This approach homogenized their ranges and facilitated comparison of their effects based on regression coefficients.

Statistical analyses were performed using the multi-model inference method (Burnham & Anderson 2002). This method allows inference to be made on the basis of all models from an *a priori* set rather than based only on the best estimated model. There were 65 535 possible combinations of all of the explanatory variables. However, only 49 151 models were evaluated because we excluded models that considered the type and the amount of nitrogen inputs at the same time because of the moderate correlation between them.

Models were compared using the adjusted Akaike information criterion (AIC) (Burnham & Anderson 2002). This method allows direct comparison of the information loss of each model in relation to the estimated best model, which has the minimum AIC ($\Delta_i = \mathrm{AIC}_i - \mathrm{AIC}_{\mathrm{min}}$). Afterwards, the Akaike weight (w_i) was calculated for each model. To assess the effect of each variable, multi-model inference from the entire set of models was used, which provides the model-averaged parameter estimates and their unconditional SE weighted by their Akaike weights. The 95% confidence intervals were also computed to evaluate the breadth of the likely magnitude of their contributions. All analyses were also conducted for the in-field species richness of the rare weeds subset.

Farm γ- and β-diversity analysis

The number of fields surveyed per farm differed according to the number of organic fields managed by a farmer. Therefore, because the number of plots per field also differed, the γ- and β-diversity values at the farm level were estimated by means of a double rarefaction, selecting four random fields per farm and three random samples per field. As no hierarchical design is present at the farm level, a linear model with a Gaussian error distribution was used after confirming the normality and homoscedasticity of the residuals. Continuous explanatory variables were standardized (see In-field diversity analysis).

The process of re-sampling and fitting the models was repeated 10 000 times. We evaluated the effect of each variable on the γ- and β-diversity values of each set of species by the density distribution of the estimated coefficients for each variable. We considered that a variable might have a relevant effect when the distribution of its estimated coefficients is not centred at zero, and thus, the distribution is clearly biased towards either positive or negative values.

Statistical analyses were performed using R 3.0.0 (R Foundation for Statistical Computing, Vienna, AT) with the lme4 package (http://CRAN.R-project.org/package=lme4) for mixed-effects models.

Results

Characteristic and rare arable weed communities: overview

Overall, we recorded 65 characteristic arable weed species, 46 of which are considered rare (App. S1). Only 183 sampled plots (9.37%) did not contain any characteristic arable species. We found at least one rare species in 1162 sampling plots. The most frequent characteristic species, recorded in >30% of the plots, were *Papaver rhoeas* L. and *Polygonum convolvulus* L. However, many of the species recorded (47 of 65) were present in <5% of the plots. Our results showed that some species that are not considered rare in local flora (de Bolòs et al. 2005) were recorded less frequently than some of these rare species. This is the case for species such as *Anchusa italica* Retz. (0.25% of plots), *Galeopsis ladanum* L. subsp. *angustifolia* (Her. ex Hoffm.) Gaudin (0.25%) and *Ranunculus arvensis* L. (2.30%). Among the species considered rare, the most frequent were *Kickxia spuria* (L.) Dumort and *Galium aparine* L. subsp. *spurium* (L.) Simonk, but they were recorded in <16% of the total plots. Therefore, despite sampling only those areas preferred by characteristic and rare weed species, these species were found to be uncommon in Mediterranean dryland arable fields. The mean (±SE) characteristic species richness per field (in three plots) estimated by rarefaction was 5.93 ± 0.17, and that of rare weed species was 1.79 ± 0.07. At the farm level, the mean (±SE) characteristic γ species richness was 12.34 ± 0.64, estimated by rarefaction, and that of rare weed species was 4.68 ± 0.30.

Field diversity

Overall, the in-field species richness of characteristic arable weeds was influenced by farming practices, field physical characteristics and PAL (Table 1). However, based on the magnitude of their effects, management variables had a much stronger effect on these species than the landscape descriptors.

Regarding past (short-term) field management, years since conversion to organic farming positively affected the richness of characteristic arable species, and fields with a high proportion of cereal crops in the rotation (cereal ratio) harboured more characteristic and rare species (Table 1).

Regarding current management, the variables that had the strongest effect based on their estimated coefficients were the crop type and sowing period, type of fertilization, weed control and animal grazing. In contrast, the type of tillage did not influence richness of either characteristic or rare species (Table 1). The characteristic and rare species richness were negatively affected by sowing of annual ryegrass compared with cereal-sown fields, as well as by spring crops compared with winter crops. Fertilizer application reduced the weed diversity of these species, but primarily when slurry was applied. Although the amount of nitrogen input affected neither the characteristic arable species nor the rare species, its interaction with the crop cover had a negative effect on both groups of species. Weed control favoured the richness of characteristic species compared with lack of weed control. Among the fields with weed control, pre-sowing tillage had a positive effect on characteristic and rare arable weeds compared with harrowing. Grazing negatively affected the richness of characteristic and rare weed species. Although the magnitude of the effect was much lower than the above-mentioned variables, we also found a positive effect of crop cover on the species richness, perhaps biased by the effect of interaction with the amount of nitrogen input. Higher PAL and perimeter/area ratio were related to lower species richness of characteristic species; however, larger fields promoted richness.

Farm diversity

The γ-diversity of characteristic species richness was affected by farm management but not by landscape simplification or inter-field distances (Fig. 2). The only factor determining total characteristic species richness at the farm level was the ratio of cereal crop fields within a farm. The γ-diversity of rare arable species richness was not influenced by factors operating at the farm scale.

The farm β-diversity was favoured by mixed farming compared to farms without stock. The number of cereal crops sown in a farm had a negative effect on the β-diversity of characteristic species, but was not affected by the landscape variables. However, the β-diversity of the subset of rare weeds was positively affected by the distance between fields; resulting in highly variable assemblages when more distant fields were considered.

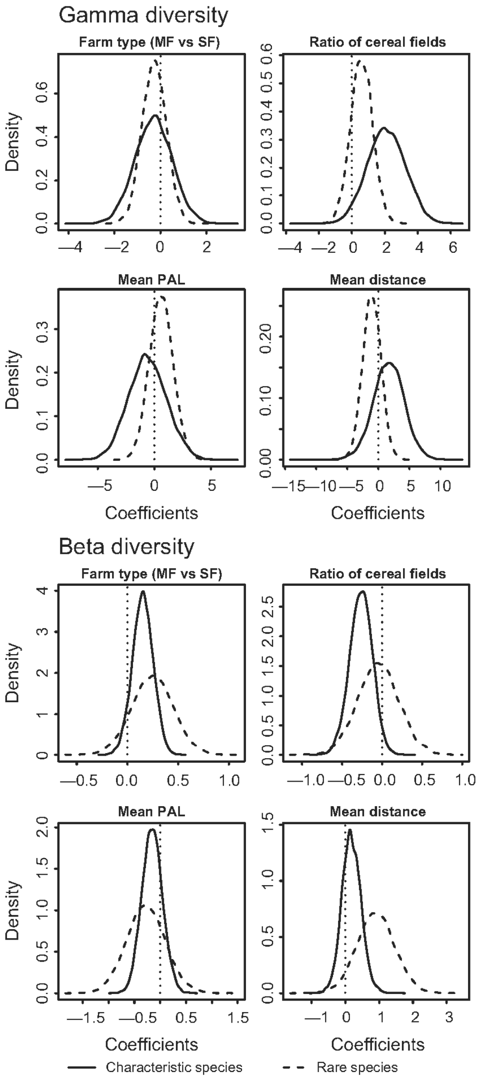

Fig. 2. Density distribution of estimated coefficients for each farm-level explanatory variable. The distribution of coefficients results from fitting the models for γ- and β-diversity of characteristic and rare weeds to 10 000 random subsets of data, and indicates the probability of a variable having an effect of a given magnitude. Farm type represents the magnitude of the difference between mixed farms (MF) and farms without stock (SF). Ratio of cereal fields, PAL (percentage of arable land) and mean distance (mean distance between fields of a farm) represent the magnitude of the slope in relation to the dependent variable.

Discussion

Characteristic and rare arable weeds surveyed

Due to the large number of fields surveyed in this study, and because the study focused exclusively on crop edges of organic fields where most characteristic arable weeds thrive, we found more characteristic weeds (65) than other studies from the same area (Romero et al. 2008; José-María et al. 2010). However, most of these species were found in very few samples: more than 70% of characteristic arable weeds had frequencies <5%, including some that are not considered rare by de Bolòs et al. (2005). This indicates the vulnerability of these species, which are very scarce even in those habitats found to be most appropriate for their growth (José-María et al. 2010).

Effects of management on in-field species richness

Most management variables affected the in-field arable species richness. Although above-ground weed flora depends largely on the specific conditions generated by the standing crop (Hawes et al. 2010), our results show that the flora was also influenced by short-term historical management. For instance, old organically managed fields harbour a higher number of characteristic weed species compared with recently converted fields. Thus, despite variation in the intensity of its practices (Clough et al. 2007; Armengot et al. 2011), organic farming seems to mitigate the decline of characteristic weed populations. A high proportion of cereals in the rotational scheme promoted the occurrence of these weeds, as previously noted (Kolářová et al. 2013), because this cropping system provides the conditions to which they are adapted. For instance, most characteristic species germinate in autumn, which matches the typical cycle of winter arable crops (Saatkamp et al. 2011). Thus, ploughing before spring sowing removes the emerged seedlings (Critchley et al. 2006), leading to a decrease in the total diversity of characteristic weeds, as observed here.

Arable weeds and crop species compete for the same resources and, hence, tend to be exclusive (Ponce et al. 2011). Characteristic arable species are usually less competitive than crops (Critchley et al. 2006), and thus, we found that they are negatively affected by farming practices favouring crop growth. This is illustrated in our study by the negative effect of fertilization on characteristic species richness, as previously described (Gabriel et al. 2005; Storkey et al. 2012). This effect is even stronger when slurry is applied because slurry generally contains higher and more labile forms of nitrogen than other fertilizer sources (Romanyà et al. 2012). The negative effect of fertilization on weed species is usually indirect because fertilizer stimulates crop growth, thus reducing light penetration and consequent weed growth (Pyšek & Leps 1991).

At the edges of fields, where we performed our surveys, sowing is less homogenous than in the centre (Romero et al. 2008). Thus, crop cover represents the actual amount of the cereal crop competing with the weeds. Its effect on characteristic and rare species richness was modulated by its interaction with the amount of available nitrogen. While this interaction is significant, the main effects of the variables involved (amount of nitrogen and crop cover) are of little interest. This significant interaction indicated that at high values of crop cover, weed species richness declined as fertilizer levels increased because some characteristic weeds are out-competed by the crop (Pyšek & Leps 1991). However, under low crop cover, characteristic and rare species are favoured by the improved resource availability without being subject to competitive pressure from the crop.

Not all management practices aimed at improving crop conditions have consistently negative effects on characteristic species. Pre-sowing weed control, such as tillage practices, favoured characteristic species richness, contrary to some findings (Santín-Montanyá et al. 2013). Our study focused on characteristic arable species adapted to annual soil disturbance (Critchley et al. 2006) and not on overall weed diversity, which may respond differently. In contrast, post-emergence mechanical weed control reduces species richness because it directly affects the established community. Similarly, characteristic species were negatively affected by animal grazing, which might represent a change to the disturbance regime other than ploughing or harvesting (Critchley et al. 2006).

Effects of landscape on in-field species richness

Despite the significant influence of landscape variables on characteristic and rare species richness, effects of such variables were of minor importance. Moreover, patterns in the response of species richness in both sets of species in relation to landscape simplification differed slightly. Simple landscapes (with high PAL) had a negative effect on characteristic species richness, but benefited the rare weeds subset. This effect on weed species richness has previously been reported (Gabriel et al. 2005; Roschewitz et al. 2005; José-María et al. 2010) and related to the relevance of alternative habitats in the surrounding landscape as a refuge for weed species, which would not be the case for characteristic weeds that preferentially thrive in arable habitats. Rare species at the crop edges were favoured by more available habitat (higher PAL), contrary to evidence from other studies (Kovács-Hostyánszki et al. 2011; Solé-Senan et al. 2014). Our results indicate that characteristic arable weeds, especially rare weeds, respond more to local conditions and thus depend more on the fields themselves than on landscape characteristics.

At the field level, larger fields held more characteristic and rare weed species, even when the effect of increased sample size was removed through rarefaction. Larger fields might support a more diverse weed flora by containing a broader range of microenvironments while still enabling intra-field exchange of diaspores. Moreover, large fields are often created from the historical amalgamation of smaller fields, which may enlarge local species pools (Marshall 2009). Nevertheless, some authors found either no effect of area on species richness (Marshall 2009) or even a negative effect (Gaba et al. 2010). These characteristic weed species thrive preferentially on field edges; thus we expected higher characteristic species richness in fields with a higher perimeter/area ratio (Gabriel et al. 2005). However, our data support the opposite effect, most likely due to the strong positive effect of area.

Effects of farming and landscape on farm diversity

A higher number of cereal crop fields within a farm led to more homogenous management among fields, which reduced the β-diversity of characteristic species richness. However, total species richness (γ-diversity) was higher in farms with a higher proportion of cereal fields. These results indicate that the higher γ-diversity was a result of higher in-field diversity found in the cereal fields instead of differences between weed communities in the fields. Regarding the different purpose of crops (e.g. hay, silage, seeds), farming practices in mixed farms are usually more heterogeneous than in farms without stock, which was reflected in the higher β-diversity for both characteristic and rare species. However, the γ-diversity of mixed farms remains equivalent to that of farms without stock.

In contrast to other studies (Baessler & Klotz 2006; Gabriel et al. 2006; Solé-Senan et al. 2014), our results showed that landscape simplification at the farm level did not have an effect on either γ- or β-diversity. This result indicates that characteristic and also rare arable species were not only more sensitive to landscape variables at the field than at the farm level but were even more sensitive to specific management practices. The β-diversity of rare arable species depended on the distance between fields. Rare weed species appear under certain local conditions, which depend on small-scale effects and on the history of the fields.

Implications for conservation

Our results show that most characteristic species are very uncommon in the sampled fields, despite being surveyed only in their preferred areas. Most species were found in very few samples, including some species that are not considered regionally rare (de Bolòs et al. 2005); therefore, the rarity of characteristic species should be revised to determine appropriate conservation strategies. Management pressure imposed on agricultural land, even under organic systems, affected characteristic arable species richness similarly to the effect on rare weeds. Implementation and maintenance of organic management guidelines could lead to the preservation of characteristic and rare arable species at field edges. However, organic farming is currently insufficient to counteract the current critical conservation status of rare weeds. Thus, it is necessary to adopt specific management practices, in addition to encouraging conversion to organic farming. As our results show, the promotion of cereal crops should be encouraged, particularly autumn-sown varieties, and slurry fertilisation and grazing should be limited to benefit both characteristic and rare species in organically managed fields.

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Supporting Information

Additional Supporting Information may be found in the online version of this article:

Appendix S1. Characteristic arable species recorded.

Supporting Information to the paper

Rotchés-Ribalta, R. et al. Both farming practices and landscape characteristics determine the diversity of characteristic and rare arable weeds in organically managed fields. *Applied Vegetation Science*

Appendix S1: List of characteristic arable species surveyed, belonging to the order *Secalietalia cerealis* Br.-Bl. 1936. Rarity according to de Bolòs *et al*. (2005) is listed as: cc (very common), c (common), r (rare), rr (very rare) and rrr (extremely rare).

Species	**Rarity**
Adonis annua L.	r
Adonis flammea Jacq.	r
Agrostema githago L.	r
Ajuga chamaepitys (L.) Schreber	r
Ammi majus L.	rr
Anchusa italica Retz.	c
Anthemis cotula L.	r
Asperula arvensis L.	r
Avena fatua L.	rrr
Bifora radians M. Bieb.	rrr
Bifora testiculata (L.) Spreng.	r
Biscutella auriculata L. subsp. *auriculata*	r
Bromus secalinus L.	r
Bupleurum rotundifolium L.	rr
Caucalis platycarpos L.	c
Centaurea collina L. subsp. *serratulifolia* (Sennen et Pau ex Hayek) Hayek	r
Centaurea cyanus L.	r
Chrozophora tinctoria (L.) Raf.	rr
Coronilla scorpioides (L.) Koch	c
Delphinium peregrinum L.	r
Delphinium pubescens DC.	r
Euphorbia falcata L.	c
Fumaria densiflora DC.	rr
Galeopsis ladanum L. subsp. *angustifolia* (Ehr. Ex Hoffm.) Gaudin	c
Galium aparine subsp. *spurium* (L.) Simonk	r
Galium tricornutum Dandy	c
Gladiolus italicus Mill.	rr
Glaucium corniculatum (L.) J.H. Rudolph	rr
Hypecoum pendulum L.	rr
Hypecoum procumbens L. subsp. *grandiflorum (Benth.)* Pau	c
Kickxia elatine (L.) Dumort.	r
Kickxia spuria (L.) Dumort.	r
Lathyrus aphaca L.	c
Lathyrus cicera L.	c
Legousia hybrida (L.) Delarbre	rr
Lithospermum arvense L. subsp. *arvense*	c
Malcolmia africana (L.) R. Br.	r
Matricaria recutita L.	r
Neslia paniculata (L.) Desv. subsp. *thracica* (Velen.) Bornm.	r
Nigella gallica Jord.	r
Papaver argemone L.	rr
Papaver dubium L.	r
Papaver rhoeas L.	cc
Papaver hybridum L.	r
Polycnemum arvense L.	rr
Polygonum convolvulus L.	c
Ranunculus arvensis L.	c
Raphanus raphanistrum L.	r
Rapistrum rugosum (L.) All.	c
Roemeria hybrida (L.) DC.	r
Scandix pecten-veneris L. subsp. *pecten-veneris*	c
Silene conoidea L.	rr
Silene muscipula L.	r
Sinapis arvensis L.	rr
Sisymbrium orientale L. subsp. *orientale*	r
Torilis nodosa (L.) Gaertn.	c
Turgenia latifolia (L.) Hoffm.	rr
Vaccaria hyspanica (Mill.) Rauschert	rr
Valerianella coronata (L.) DC.	rr
Vicia benghalensis (L.)	r
Vicia bithynica (L.) L.	r
Vicia peregrina L.	c
Vicia villosa subsp. *varia* (Host) Corb.	r
Viola tricolor L. subsp. *arvensis* (Murray) Gaudin	c

Capítol 2

Which conditions determine the presence of rare weeds in arable fields?

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RESUM

Quines condicions determinen la presència de les espècies segetals rares en els camps cerealistes de secà?

La intensificació de la gestió agrícola en les últimes dècades ha provocat que algunes espècies arvenses especialistes dels camps cerealistes de secà (o espècies segetals) hagin esdevingut rares o fins i tot localment extingides. La conservació d'aquestes espècies, però, és complicada ja que el seu hàbitat preferent és dedicat a la producció de cultius i, precisament, les espècies arvenses han estat considerades com un impediment per a aquesta producció. Tot i això, és convenient dedicar esforços a la conservació de les espècies segetals ja que són un component important de la biodiversitat dels sistemes agrícoles i proporciona més diversitat funcional i estructural a aquests hàbitats.

En realitat, però, és difícil elucidar quins són els efectes de les pràctiques agrícoles sobre aquestes espècies més rares, ja que apareixen de manera molt esporàdica. Amb aquest estudi, hem superat parcialment les limitacions causades per les baixes freqüències mitjançant l'anàlisi de la seva presència en un gran nombre de punts de mostreig (1957) a les vores de 304 camps ecològics, el que ha permès maximitzar-ne la detecció. Es van prendre aquestes dades per tal de 1) determinar els efectes de les pràctiques agrícoles i de les condicions locals en la presència d'espècies arvenses rares als camps i 2) avaluar les similituds entre les respostes d'algunes espècies arvenses rares seleccionades a aquestes variables.

Es van mostrejar les espècies arvenses característiques dels camps cerealistes de secà (ordre fitosociològic *Secalietalia cerealis* Br.-Bl. 1936) en diferents punts de mostreig separats entre ells 80 m a les vores dels camps ecològics. A cada punt de mostreig se'n va avaluar la presència en una àrea de 2 × 5 m. Es van considerar les espècies rares segons la categorització que en fan de Bolòs et al. (2005). Es van avaluar els efectes de la gestió agrícola dels camps, així com de les característiques descriptives dels camps i del paisatge sobre la freqüència d'aquestes espècies rares per camp mitjançant un model mixt generalitzat amb un terme d'error de distribució binomial. A més, a escala local (punt de mostreig) es va avaluar l'efecte del recobriment del cultiu i del recobriment de les espècies arvenses totals i el tipus de marge adjacent, a part de les variables considerades a escala de camp, sobre la probabilitat de presència d'aquestes espècies mitjançant el mateix tipus de model estadístic. Es va calcular el coeficient de determinació (R^2) considerant tant els factors fixes i aleatoris com només els factors fixes. Es van avaluar, també, els efectes dels diferents factors sobre la presència de cadascuna de les espècies més freqüents

individualment. La informació dels models individuals es va combinar mitjançant una meta-anàlisi, per tal de determinar patrons de resposta generals en les tendències individuals.

Es van trobar 46 de les 65 espècies segetals rares que poden aparèixer a la zona d'estudi, però més de la meitat de les espècies va aparèixer en menys de l'1 % dels punts de mostreig. El cultiu de cereals, tant sol com barrejat amb lleguminoses, va afavorir la presència d'espècies arvenses rares als camps, ja que determina les condicions a les quals aquestes espècies estan adaptades (Kolářová et al. 2013). L'ús de fertilitzants determina unes condicions menys favorables per a aquestes espècies, ja que estimula el creixement del cultiu, més competitiu i eficient a l'hora de captar els nutrients, en detriment de les espècies arvenses que tenen menys capacitat competitiva (Storkey et al. 2012). A les vores dels camps, però, es mantenen les poblacions d'espècies segetals i també de les que són rares quan es donen unes condicions favorables per al creixement del cultiu ja que aquest últim no sol presentar recobriments gaire alts. En els camps més grans la probabilitat de presència d'espècies arvenses rares és més alta, ja que aquests camps probablement tenen el seu origen en l'amalgamació de camps més petits, fet que incrementaria el conjunt local d'espècies (Marshall 2009).

Ara bé, una proporció important de la variància en la presència d'espècies arvenses rares va ser explicada pels factors aleatoris relacionats amb la finca i amb el camp. Això indica que els efectes estocàstics (no controlats en el mostratge) tenen força importància a l'hora de determinar la presència d'aquestes espècies en un determinat camp d'una finca.

Tot i que les espècies individuals no van respondre de manera significativa a la majoria de factors testats, amb la meta-anàlisi es van detectar uns patrons de resposta comuns significatius. Les espècies individuals van tendir a aparèixer preferentment en camps de cereals sembrats a la tardor després d'una llaurada sense inversió del sòl, condicions que proporcionen els requeriments per a la germinació d'aquestes espècies particulars (Gruber & Claupein 2009; Saatkamp et al. 2011).

Malgrat que moltes espècies segetals són manifestament rares, moltes d'elles s'han detectat en el mostratge de camp. Aquest fet dóna certa esperança pel que fa a la seva conservació. Ara bé, la seva presència es troba poc determinada per la gestió i, en canvi, sembla dependre força de la història particular de cada finca o camp. Així doncs, per a la conservació d'aquestes espècies s'haurien de promoure les pràctiques agrícoles específiques que les afavoreixen, com la sembra de cereal, especialment de tardor, o una fertilització reduïda, però en aquelles zones on les espècies rares encara són presents per així evitar la seva pèrdua.

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Which conditions determine the presence of rare weeds in arable fields?

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ABSTRACT

The intensification of agricultural management has caused some weed species to become rare in arable farming systems. It is difficult to disentangle which management practices are the least harmful for the conservation of rare arable weeds because of their sparse presence. In this research, we overcame the limitations of previous analyses of rare weeds by analyzing them in a large number of plots (1957) at the edges of multiple organic fields (304), which maximized the probability of detecting these species. We evaluated the relationships between farming practices and local site conditions and the presence of rare arable species that are characteristic of cereal fields.

We detected 46 of the 65 rare weeds that are known to inhabit the study area, but their frequency was very low. Cereal crops, either alone or in mixtures with legumes, enhanced the probability of finding rare weed species, while fertilization had a detrimental effect. Other management practices that were considered had no effect on the presence of rare arable weeds. However, selected rare species tended to fare better under particular local conditions and to be favored by specific management practices. In contrast, a significant amount of the variance of the rare weed presence was explained by farm-related and field-related random factors. Thus, the occurrence of rare arable species is apparently determined by stochastic factors that may be related to the local species pool that likely depends on the history of fields and farms. Therefore, conservation efforts should be focused on areas currently inhabited by rare arable species.

1. Introduction

Conserving arable weed species is somewhat problematic because their preferred habitat, arable fields, is primarily devoted to crop production (Fried et al., 2009). Increases in farming efficiency to enhance productivity have resulted in fields becoming less diverse, with few non-crop plants tolerated (Robinson and Sutherland, 2002). Agriculture has repeatedly been identified as one of the main causes of biodiversity loss worldwide (Elsen, 2000; Rich and Woodruff, 1996; Storkey et al., 2012). This is primarily due to the large cropland areas that are devoted to grow crops, which diminish non-crop habitats such as hedgerows and field margins, and the widespread use of pesticides and fertilizers (McLaughlin and Mineau, 1995). Therefore, conserving farmland biodiversity requires less intense farming practices (Tscharntke et al., 2005).

In recent decades, many arable weed species have suffered such a critical population decline that they have become rare or even locally extinct in many countries (Baessler and Klotz, 2006; Cirujeda et al., 2011; Fried et al., 2009; Storkey et al., 2012). Many of these species are found in the Red Data Lists of some European countries (Cheffings and Farrel, 2005; Kleijn and van der Voort, 1997 Türe and Böcuk, 2008), which constitutes the basis for most conservation strategies (Aboucaya et al., 2000; Kleijn et al., 2006). However, in Spain and other countries in the Mediterranean area, rare arable weed species are not included in the Red Data Lists or in conservation plans because these species are considered non-native and dependent on the maintenance of artificial habitats (Sáez et al., 2011). Conservation of rare arable species is crucial because of their intrinsic value as both components of biodiversity and key indicators of traditional and low-intensity agriculture. In addition, these rare species provide a greater variety of forms, compositions and functions than do the few crop species that dominate arable land and constitute a valuable resource for

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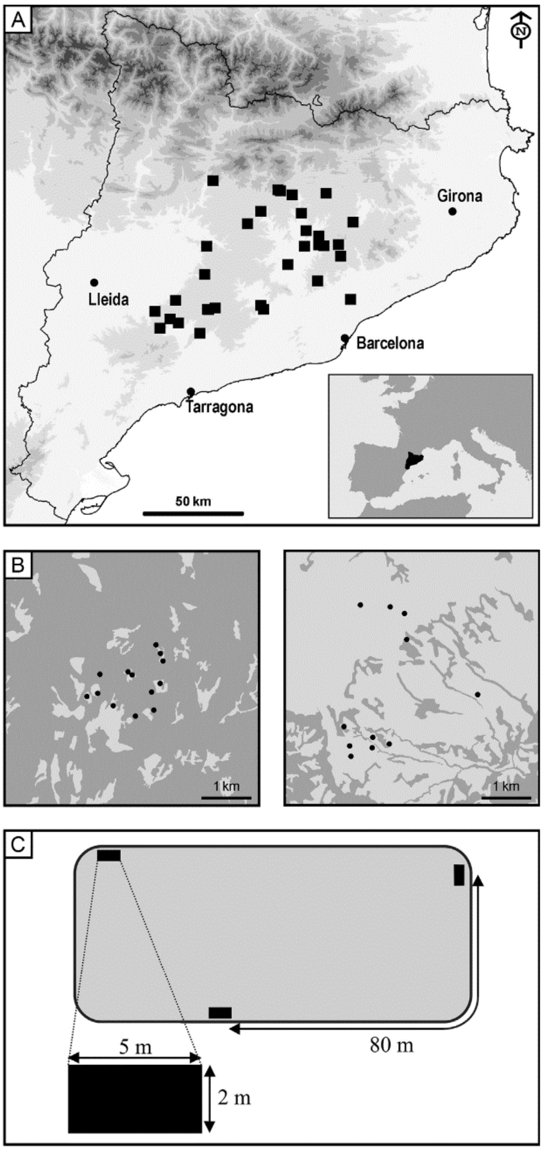

Fig. 1. (A) Locations of the 32 farms (squares) within Catalonia (NE Spain). Gray tones indicate elevation contours of 500 m. (B) Fields sampled (dots) at two farms taken as examples of complex (left) and simple (right) landscapes. Light gray represents arable habitats while dark gray includes other natural and semi-natural habitats. (C) Plots of 2 m × 5 m size spaced 80 m apart from each other were surveyed within each field.

pollinators, herbivores, predators and parasitoids (Caballero-López et al., 2010; Hawes et al., 2003; Tscharntke et al., 2005).

The distribution of arable weed species is determined by many parameters, among which history of land management and landscape composition, weather conditions, seed dispersal and other stochastic factors can play an important role (Ryan et al., 2010). Nevertheless, it is widely acknowledged that less intensive farming practices, such as those used in organic farming where herbicide and chemical fertilizer inputs are banned, tend to be beneficial for the richness and diversity of arable species (Cirujeda et al., 2011; Gibson et al., 2007; Kleijn et al., 2009), as well as the occurrence of rare arable weeds (Romero et al., 2008). However, there is considerable variation in management intensity among organic farms (Armengot et al., 2011a; Clough et al., 2007). This leads to highly versatile and seemingly contradictory effects of organic vs. conventional farming on diversity (Bengtsson et al., 2005). Thus, it may be appropriate to consider particular management practices to evaluate their impact on the presence of rare arable species.

Several studies have shown how weed species diversity is determined by the surrounding landscape, which acts as a refuge and source of propagules that can colonize crops (Gabriel et al., 2005; José-María et al., 2010; Poggio et al., 2010; Roschewitz et al., 2005; Solé-Senan et al., 2014). For this reason, it is important to consider the composition and amount of natural and semi-natural habitats that are adjacent to fields and landscape structure as factors that may contribute to variations in weed diversity.

The purpose of our research was to determine which field and local conditions are more suitable for the occurrence of rare arable species and to outline appropriate management practices that may promote their conservation. The specific objectives of this study were (1) to assess the effects of farming practices and local conditions on the occurrence of rare weed species within fields and (2) to investigate similarities between the responses of selected species to these variables. This study overcomes limitations of analysis that are related to low frequencies and usually affect studies of rare arable species, by surveying them in many plots at the edges of multiple organic fields where those species are most likely found (José-María et al., 2010; Kovács-Hostyánszki et al., 2011). Thus, this sampling method maximizes the probability of detection of each rare species (Thompson, 2004).

2. Material and methods

2.1. Study site

The sampling was conducted in 2011 in Catalonia, northeastern Spain (41°22′–42°06′N; 0°59′–2°12′E). We selected organically managed fields from 32 farms within an area spanning 100 km × 80 km (Fig. 1). The average (±standard error) altitude of the surveyed sites is 558 m a.s.l. (±30 m and ranging from 95 to 871 m a.s.l.). The fields have basic soils with loamy and clayish textures. The climate is Mediterranean, with mean annual temperatures of 12.6 ± 0.2 °C and an average precipitation of 637 ± 21 mm (Ninyerola et al., 2005). Average monthly temperatures are always positive, but frost can occur from December to February, and is normally restricted to a few hours per day. Natural habitats in the study area include pine (*Pinus halepensis* Mill. and *P. nigra* Arnold) and oak (*Quercus ilex* L. and *Q. faginea* Lam.) woodlands, shrublands, small stands of perennial-dominated grasslands, and riverine vegetation.

2.2. Plant survey

A total of 304 organically managed fields were surveyed during May and June 2011 (just before harvest). The selected fields had been sown in the previous growing season with the annual crops that are usually included in the winter cereal crop rotation (small grain cereals, legumes, ryegrass and crop mixtures containing cereals and legumes). To maximize the detection of rare arable weeds, the weed survey was restricted to the edges of the fields, which are defined as the first cultivated meters adjacent to field margins (Marshall and Moonen, 2002). The sampling plots (2 m × 5 m) were established 80 m apart at the edges of the fields

Table 1
Characterization of management, field and landscape descriptors and the local conditions of the sampled farms ($n = 32$), fields ($n = 304$) and plots ($n = 1957$). The mean and range (in brackets) of the continuous variables and the proportion of fields and farms with the stated practices for the discrete variables are shown.

Field management	
Years from conversion to organic management	9.67 [1,25]
Cereal ratio[a]	0.56 [0,1]
Soil tillage	
No till and no inversion tillage	199/304
Inversion tillage	105/304
Current crop	
Cereal	170/304
Mixture	87/304
Legume	17/304
Ryegrass	30/304
Seed origin	
Reuse seeds	192/304
Purchased seeds	112/304
Sowing time	
Autumn sowing	246/304
Spring sowing	58/304
Sowing density (kg ha^{-1})	154.81 [16.5, 384]
Type of N inputs	
No fertilization	119/304
Manure	167/304
Slurry	18/304
Amount of N inputs (kg ha^{-1})[b]	47.44 [0600]
Weed control	
No control	176/304
Tillage (pre-sowing control)	89/304
Harrowing (post-sowing control)	39/304
Animal husbandry	
No grazing	213/304
Grazing	91/304
Farm management	
Farm type	
Stockless farms	11/32
Mixed farms	21/32
Field and landscape descriptors	
Percentage of arable land (PAL)	46.58 [6.83, 100]
Field area (ha)	1.54 [0.16, 15.04]
Field shape (perimeter/area)	0.06 [0.01, 0.19]
Local conditions	
Crop cover (%)	51.97 [0100]
Weeds cover (%)	32.82 [0100]
Habitat of the adjacent margin	
Ruderal vegetation	813/1957
Grasslands	531/1957
Woody vegetation	517/1957
Other habitats	96/1957

[a] Calculated as the proportion of cereal crops in the 5-year rotational scheme of each field.

[b] Calculated using local tables of N content (Campos Pozuelo et al., 2004).

(Fig. 1). Thus, depending on the field size, a minimum of three and a maximum of ten plots were sampled per field. In each plot, we recorded the presence of rare arable weeds. These were defined as characteristic species of the phytosociological order *Secalietalia cerealis* Br.-Bl. 1936 (communities of dryland winter cereal fields in a Mediterranean climate on basic soils) that are considered rare in the study area (de Bolòs et al., 2005) (Table A1). These species are categorized into rare (r), very rare (rr) and extremely rare (rrr) according to their frequencies and abundances in relevés from various published sources and in herbarium collections. The nomenclature of plant species and their attribution to the phytosociological order follows that of de Bolòs et al. (2005). In each plot, we also recorded the habitat type of the adjacent margin and crop and total weed cover visually.

2.3. Explanatory variables considered

Farmers were interviewed to obtain information about their farming practices on each sampled field and farm during the 5 years preceding the sampling (Table 1). The selected variables reflect recent field management (number of years elapsed since conversion to organic farming and the cereal ratio, which is the proportion of cereal crops in the five-year rotational scheme) and current management during the period 2010–2011 (all other variables) of each field. The presence (mixed) or absence of livestock was used as a management variable at the farm level.

Some field descriptors, such as the area of the field and the shape of the field (perimeter-to-area ratio), were included in the model as covariates that may determine the presence of rare arable weeds in the fields. The percentage of arable land characterized within a 1 km radius around each field was considered as a landscape descriptor. We used the Catalan Habitats Cartography (Carreras and Diego, 2004) to calculate the percentage of arable land.

Local conditions at the sampling plot were also considered as variables that may determine the presence of rare arable species. These variables were the habitat type of the adjacent margin and the percentage cover of crop and weeds. Adjacent margins were categorized into ruderal vegetation, grasslands, woody vegetation (including shrublands and woodlands) and other habitats found in very low frequencies (including bare soil, arable land and wet ditch vegetation) (Table 1).

2.4. Analyses of the occurrence of rare weeds

The study was conducted at both the field ($n = 304$) and plot ($n = 1957$) levels. At the field level, we evaluated the probability of finding rare arable species in a field using the proportion of sampled plots per field that contained rare species. The effect of all recorded management variables (Table 1) on the probability of the presence of rare species in the field was analyzed using generalized mixed-effect models with a binomial error distribution, including farm as a random factor. The values of the response variable were included as binomial data using the number of sampled plots that contained rare species and the number of plots in which rare species were absent. Therefore, the total number of plots in each field was used to weight the probability that a rare species occurs in a field. Continuous explanatory variables were standardized (by subtracting the mean and dividing by their standard deviation) to homogenize their ranges of variation and facilitate the comparison of their effects based on regression coefficients. For the type of nitrogen inputs and weed control variables, orthogonal contrasts were fixed a priori to compare the different levels in a meaningful way (see Table 2). Because we did not find strong correlations between the explanatory variables, all of them were included in the model without interactions to carry out a general exploration of the rare arable species behavior.

At the plot level, we analyzed the probability of the presence of rare arable species within the plots at the field edges. Data on the presence of at least one rare species were evaluated to test the effects of local conditions (Table 1) using generalized mixed-effects models. Field and farm management variables, as well as field and landscape descriptors, were also considered in this model. We included farm and field (nested within farm) as random factors. Because the presence data were entered as binary information, we used a model with a binomial error distribution. Continuous explanatory variables were also standardized and, again, orthogonal contrasts were fitted for the type of nitrogen inputs and weed control variables.

To assess the goodness of fit of the models, we computed their coefficient of determination (R^2). These coefficients were expressed as the marginal R^2, accounting for the variance

Table 2
Effects of the field and farm management and field and landscape structure variables on the occurrence of at least one rare arable species at the field level. Estimated coefficients and their standard errors for the linear mixed models, degrees of freedom for each variable (DF) and *P*-values (*** when *P*-value < 0.001; ** < 0.01; * < 0.05 and · < 0.1) are shown.

	Estimate ± SE	DF	*P*-values
Field and farm management			
Years from conversion	−0.087 ± 0.148	254	0.559
Cereal ratio[a]	0.188 ± 0.370	254	0.612
Inversion (vs. no inversion) tillage	0.139 ± 0.335	254	0.678
Current crop			
Mixture (vs. cereal)	−0.104 ± 0.203	254	0.607
Legume (vs. cereal)	−0.897 ± 0.302	254	0.003**
Ryegrass (vs. cereal)	−1.407 ± 0.316	254	0.000***
Reuse (vs. purchased) seeds	−0.237 ± 0.228	254	0.299
Spring (vs. autumn) sowing	−0.295 ± 0.224	254	0.189
Sowing density[b]	0.121 ± 0.094	254	0.202
Type of N inputs			
Fertilized (vs. no fertilized)	−0.295 ± 0.152	254	0.052·
Slurry (vs. manure)	−0.296 ± 0.240	254	0.218
Amount of N inputs (log (N + 1))	0.307 ± 0.200	254	0.124
Weed control			
Control (vs. no control)	−0.065 ± 0.076	254	0.394
Tillage (vs. harrowing)	−0.038 ± 0.214	254	0.859
Grazing (vs. no grazing)	−0.242 ± 0.194	254	0.212
Mixed farms (vs. stockless farms)	−0.264 ± 0.422	30	0.532
Field and landscape descriptors			
Field area	0.214 ± 0.070	254	0.002**
Field shape (perimeter/area)	−0.025 ± 0.074	254	0.733
Percentage of arable land (PAL)	0.166 ± 0.119	254	0.164

Model deviance = 1231.1.

[a] Calculated as the proportion of cereal crops in the 5-year rotational scheme of each field.

[b] Standardized for each crop type.

explained by the fixed factors, and the conditional R^2, which is the variance jointly explained by both fixed and random factors (Nakagawa and Schielzeth, 2013).

2.5. Analyses of the occurrence of selected rare arable species

Analyses to assess the effects of both field and plot variables on the presence of each species were also performed using mixed-effects models with binomial errors. We considered only those rare species with more than 19 occurrences, which represents 1% of the sampled plots. This yielded 19 rare arable species. These analyses allowed us to ascertain whether the pattern of presence found for the pool of rare weeds responded to consistent patterns among species.

Table 3
Effects of local conditions on the presence of rare characteristic arable species sampled at the plot level. Estimated coefficients and their standard errors for the linear mixed model, the degrees of freedom for each variable (DF) and the *P*-values (*** when *P*-value < 0.001; ** < 0.01; * < 0.05 and · < 0.1) are shown. Field management practices and landscape structure were also included in the model but are not shown in the table.

	Estimate ± SE	DF	*P*-value
Local conditions			
Crop cover	0.196 ± 0.085	1648	0.022*
Weeds cover	0.380 ± 0.085	1648	0.000***
Habitat of adjacent margin			
Grasslands (vs. ruderal vegetation)	−0.314 ± 0.321	1648	0.329
Woody vegetation (vs. ruderal vegetation)	−0.050 ± 0.161	1648	0.756
Other habitats (vs. ruderal vegetation)	0.153 ± 0.167	1648	0.359

Model deviance = 2217.3.

Moreover, we carried out a meta-analytic approach to combine the information from individual species' models of the 19 most frequent rare arable species based on the *P*-values of the estimated effects (Zaykin, 2011). The combination of *P*-values can be used to support a common hypothesis that has been tested in several studies (in our case, several species). We used the weighted Z-test, which is Stouffer's method (also known as the 'inverse normal' test) (Zaykin, 2011). For each variable, we used the inverse of the coefficient's estimated standard errors as weights, as recommended in Zaykin (2011). Two-sided *P*-values are generally inappropriate for the meta-analytic combination of *P*-values. Therefore, the individual *P*-values were converted to one-sided *P*-values before combination as follows: $P_{\text{one-sided}} = P_{\text{two-sided}}/2$ if the effect direction (either positive or negative) was the same as expected, and $P_{\text{one-sided}} = 1 - P_{\text{two-sided}}/2$, otherwise. Once they were combined, the results were converted back to two-sided as follows: $P_{\text{two-sided}} = 2P_{\text{one-sided}}$ if $P_{\text{one-sided}}$ is lower than 0.5 and $2(1 - P_{\text{one-sided}})$ otherwise.

For the analyses, we used R 3.0.3 (R Development Core Team, 2013) with the lme4 package (Bates et al., 2014) for the generalized mixed models and the MuMIn package for the R^2 calculation (Bartoń, 2013).

3. Results

3.1. Overview of rare arable weed communities

Overall, we recorded 46 characteristic arable species that are considered to be rare by de Bolòs et al. (2005). These species are included in a list of 65 rare arable species with a distribution area that matches the study area (Table A1, Supplementary data). This means that we were able to detect more than 70% of the rare arable weed species that may occur in the study area. We found at least one rare species in 1162 plots from the total of 1957 sampled plots. Nevertheless, most of the rare species (27 of 46) occurred in less than 1% of the surveyed plots, and many of them were present in only one or two plots. The most frequent species were *Kickxia spuria* (L.) Dumort. and *Galium aparine* subsp. *spurium* (L.) Simonk, and even these were recorded in less than 16% of the plots.

3.2. Rare arable species at the field level

The presence of rare arable weeds was significantly affected only by a few current management practices (Table 2). Cereal crops encouraged the presence of rare weeds belonging to the *Secalietalia cerealis* order (i.e., typical of winter cereal fields) more than legume or ryegrass crops. Fertilization had a slight negative effect on the occurrence of rare arable species. Larger fields tended to have a higher probability of sustaining rare weeds.

The fraction of the variance explained by the random factor farm was larger than that explained by the fixed factors. Conditional R^2 (0.28), which accounts for the variance explained jointly by fixed and random factors, was higher than the marginal R^2 (0.10) or the variance explained by all of the fixed factors. This difference indicates that the fraction explained by the random effects that depend on farm was more important.

3.3. Rare arable species at the plot level

The percentage of crop cover and especially of weed cover showed a significant positive relation with the presence of rare arable species (Table 3). On the contrary, the type of habitat in adjacent boundaries did not influence the presence of rare arable species. Among the field-level variables included in the plot-level

Table 4
Number of species with positive (+) or negative (−) effect of each variable considered in the mixed-effect models for the presence of each rare arable species analyzed. The number of species with a significant effect is in parentheses. 'Combined *P*-value' represents the *P*-values of Stouffer's meta-analytic approach used to combine the information of each particular species model. The following notation is used for size of the *P*-values: *** when *P*-value < 0.001; ** < 0.01; * < 0.05 and · < 0.1.

	+	−	Combined *P*-value
Field and farm management			
Years from conversion	17 (6)	2 (0)	0.000***
Cereal ratio^a	14 (0)	5 (0)	0.009**
Inversion (vs. no inversion) tillage	2 (0)	17 (1)	0.000***
Current crop			
Crop mixtures (vs. cereal)	9 (1)	10 (0)	0.281
Legume (vs. cereal)	7 (0)	12 (1)	0.044*
Ryegrass (vs. cereal)	2 (0)	17 (6)	0.000***
Reuse (vs. purchased) seeds	7 (0)	12 (0)	0.163
Spring (vs. autumn) sowing	0 (0)	19 (3)	0.000***
Sowing density^b	16 (2)	3 (0)	0.000***
Type of N inputs			
Fertilized (vs. no Fertilized)	12 (0)	7 (0)	0.561
Slurry (vs. manure)	14 (0)	5 (0)	0.186
Amount of N inputs (log (N + 1))	12 (0)	7 (0)	0.597
Weed control			
Control (vs. no control)	11 (1)	8 (0)	0.12
Tillage (vs. harrowing)	6 (0)	13 (0)	0.362
Grazing (vs. no grazing)	3 (0)	16 (3)	0.000***
Mixed farms (vs. stockless farms)	13 (0)	6 (0)	0.889
Field and landscape descriptors			
Field area	15 (2)	4 (0)	0.003**
Field shape (perimeter/area)	4 (0)	15 (2)	0.001**
Percentage of arable land (PAL)	11 (1)	8 (1)	0.331
Local conditions			
Crop cover	11 (0)	8 (0)	0.222
Weeds cover	15 (3)	4 (0)	0.000***
Habitat of adjacent margin			
Grasslands (vs. ruderal vegetation)	8 (1)	11 (0)	0.26
Woody vegetation (vs. ruderal vegetation)	3 (0)	16 (1)	0.001**
Other habitats (vs. ruderal vegetation)	11 (2)	8 (0)	0.003**

[a] Calculated as the proportion of cereal crops in the 5-year rotational scheme of each field.

[b] Standardized for each crop type.

model, only ryegrass sown during the current growing season (vs. cereal-sown fields) showed a negative effect on the presence of rare species at the plot level (-1.31 ± 0.50, $P = 0.009$).

Random factors also largely influenced the presence of rare species at the plot level. The variability explained by all of the fixed factors (marginal R^2) was only 0.10. However, the conditional R^2 was 0.24 when farm was considered as the only random factor and 0.47 when farm and field were both considered as random factors, together with the fixed factors.

3.4. Patterns of presence of selected species

Analyses of the effects of both field and plot variables showed that very few of the assessed parameters significantly affected the presence of the 19 most common rare species when separately analyzed (Table 4). Only the presence of ryegrass crops, whichnegatively influenced the presence of rare species, and the years since the conversion to organic farming, which had a positive effect, affected up to six of the 19 species analyzed (Table 4). Analyses that accounted for all of the models of the 19 species revealed that weed cover at the plot level was beneficial to these rare arable species. In addition, these rare arable species were more likely to be found in larger fields that were under organic management for a longer time and that had a higher proportion of cereals in the rotation scheme. They appeared preferentially in fields sowed with cereal or crop mixtures instead of legumes or ryegrass and were not favored by spring sowing. Tillage with soil inversion and grazing after the preceding harvest determined a smaller presence of particular rare weeds. Rare weeds tended to not occur at the field edges that neighbored woody vegetation.

4. Discussion

4.1. Status of rare arable weeds

A large proportion of the rare arable species occurred at the edges of the organically managed fields. We detected more than 70% of the rare species that de Bolòs et al. (2005) listed as inhabiting the study area. This high percentage may be due to the large number of plots surveyed at the edges of fields and because the study was focused where these species are preferentially found. However, 40% of the total surveyed plots were completely devoid of rare species and, when the rare species were present they had extremely low frequencies. These data confirm the current scarcity of the studied species (Chamorro et al., 2007) while providing some hope for their conservation because many of them are still present in arable fields of Catalonia.

4.2. Presence of rare arable species in fields

One of the strongest agricultural filters on the presence of rare arable species was the standing crop type, which has also been reported in other studies (Fried et al., 2009; Marshall, 2009). Because rare arable weeds as defined in this study are specialists of winter cereal cropping systems, sowing cereals or a mixture containing cereals provides the conditions that favor the presence of these species (Kolářová et al., 2013).

Fertilized fields were marginally detrimental to rare arable weeds, as also reported by Storkey et al. (2012). Arable weeds and crop species compete for the same resources and hence tend to be mutually exclusive (Ponce et al., 2011; Critchley et al., 2006). This negative effect of fertilization on weed species usually acts indirectly by stimulating crop growth, which in turn decreases light penetration and reduces weed growth (Kleijn and Van der Voort, 1997).

The inclusion of field descriptors (field area and shape) and the proportion of arable land in the model allowed the possible effect of these parameters on the presence of these rare species to be discarded. Actually, only field areas contributed to an explanation of the presence of rare arable weeds. Larger organically managed fields were more likely to hold rare arable species than smaller fields with similar management, probably because they came from the historical amalgamation of smaller fields that may have enlarged the local species pool (Marshall, 2009). The presence of rare weeds may be primarily determined by the local species pool that is maintained by the buffer effect of the soil seedbank (Hiltbrunner et al., 2008).

4.3. Local site conditions favoring rare arable species

Despite the fact that weeds and crop species tend to be mutually exclusive (Ponce et al., 2011), both crop and weed species (including the rare ones) apparently are able to coexist at the edges of organic farming fields. This particular result may be related to the low crop cover values at the studied field edges (ca. 50% on average). At the edges of the fields, lower organic fertilizer inputs, poor soil conditions and sowing failure may strongly limit crop performance and reduce the importance of crop-weed competition (Dutoit et al., 2007; Romero et al., 2008). Thus, favorable conditions for the crop in organically managed field edges would also benefit weeds, which would in turn favor the presence of rare species.

We assumed a potential effect of the adjacent boundary on the presence of rare arable species at the field edge because, as other studies have indicated, this could act as a refuge for these species (Gabriel et al., 2005; José-María et al., 2010; Poggio et al., 2010; Roschewitz et al., 2005). Because rare characteristic arable species are found almost exclusively in arable habitats and require periodic disturbances, we expected that the adjacent habitat that could best act as a refuge would be a ruderal one. On the contrary, we also expected that more competitive vegetation at the adjacent boundary, such as woody vegetation, would be detrimental for the presence of rare arable species. However, our results indicated no effect of any of these habitats on the presence of rare arable weeds.

4.4. Species-specific responses

Overall, very few of the many parameters we considered actually influenced the presence of the particular rare arable species that were analyzed. Most studies have found that management practices exert an important influence on the presence and diversity of arable weed species in arable fields (Albrecht, 2003; Armengot et al., 2011b; José-María et al., 2010). However, the occurrence of rare arable species may instead respond to stochastic factors that have led them to appear and remain at particular sites. In light of these results, we could assume that the presence of these species and the determination of their rarity are the result of specific traits of each species (Pinke and Gunton, 2014; Storkey et al., 2010), which would be an interesting point to consider in future conservation studies.

Nevertheless, with the meta-analysis we are able to detect an overall trend of these 19 rare arable species to appear under particular conditions and be favored by specific farming practices. This highlights a similar response of these species to the variables considered. Cereal crops or mixtures containing cereals and legumes, both in the current season and at high frequencies in a rotation scheme, benefited these rare arable species because the autumn sowing time of cereals is perfectly coupled with their germination requirements (Saatkamp et al., 2011). Slight annual periodic soil disturbances without soil inversion tended to favor the presence of rare arable weeds by generating the appropriate conditions for germination of seeds without burying them into the soil (Gruber and Claupein, 2009).

These rare weeds were more likely to appear in those spots with high weed cover, indicating that suitable local conditions for weeds in general were also appropriate for the rare ones. Therefore, practices that enhance weed diversity within crop fields would also be suitable for rare arable species conservation (Romero et al., 2008). Cropland edges are prone to be negatively affected by competing neighboring vegetation because it reduces the availability of soil nutrients and light through belowground competition and shading. That is why globally the 19 rare species tended to be excluded at the crop edges adjacent boundaries that were dominated by more competitive vegetation such as woodlands and shrublands. The significant effects of the other types of adjacent boundaries must be interpreted with caution because they were represented in very few samples.

4.5. Importance of random factors

Farm and field random factors significantly increased the explained variance in the occurrence of rare characteristic weeds over the fixed factors. Field effect was more important than farm effect, but even the farm alone explained more variance than did all of the fixed factors together. The importance of the random factors suggests that the occurrence of rare species is controlled by factors other than the local conditions that were considered, field and farm management and field descriptors or landscape simplification, and depends to a large extent on the farms and fields themselves. Although we were able to find a trend of particular rare arable species to appear under certain conditions, the importance of the random factors on the overall presence of these species is evident. Therefore, the presence of rare arable species at a particular site would be determined, to a large extent, by the diversity of the seeds that remain in the soil seedbank, which is largely set by the field management history (Hiltbrunner et al., 2008).

5. Conclusions

Our data reinforce the fact that rare characteristic arable species occur at the edges of organically managed fields. Nevertheless, these species face an actual threat because they are found at very low frequencies. Therefore, the moment is right to take action for their conservation, taking into account that many of them are still present in organic arable cropping systems.

The foundations for the conservation of rare arable species must consider very particular management parameters, such as the promotion of sowing winter cereal crops and fine-tuning fertilization. Particular rare arable species showed a trend to appear preferentially when autumn sowing took place or when no inversion tillage was conducted. Nevertheless, the inconsistencies between the overall occurrence analysis and the meta-analysis reflect the difficulty in understanding the role of farming practices in explaining their conservation status. For this reason, it would be interesting to promote further research on the ecological requirements at the species level to develop appropriate conservation strategies.

The presence of rare arable species was highly explained by field and farm random factors, which are the result of the variability in the local species pools. Therefore, conservation efforts based on specific practices to favor rare weeds should focus on those areas where the species are currently settled.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.agee.2015.01.022.

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Supplementary data to the paper

Rotchés-Ribalta, R. et al. Which conditions determine the presence of rare weeds in arable fields? *Agriculture, Ecosystems & Environment*

Table A1: List of the rare arable species in the studied area, as considered by de Bolòs *et al*. (2005). Rarity is specified as: r (rare), rr (very rare) and rrr (extremely rare). The number of plots with presence of each species surveyed with respect to the total number of plots surveyed (1957) is indicated as percentage.

Species	**Rarity**	**%**	**Species**	**Rarity**	**%**
Adonis aestivalis L.	rr	-	*Kickxia elatine* (L.) Dumort.	r	2.76
Adonis annua L.	r	2.15	*Kickxia spuria* (L.) Dumort.	r	15.94
Adonis flammea Jacq.	r	0.92	*Legousia hybrida* (L.) Delarbre	rr	0.46
Agrostema githago L.	r	1.58	*Linaria hirta* (L.) Moench	rr	-
Ajuga chamaepitys (L.) Schreber	r	6.03	*Lolium temulentum* L.	rr	-
Alopecurus myosuroides Huds.	rr	-	*Malcolmia africana* (L.) R. Br.	r	0.10
Ammi majus L.	rr	1.38	*Matricaria recutita* L.	r	2.35
Anthemis cotula L.	r	9.81	*Neslia paniculata* (L.) Desv. subsp. *thracica* (Velen.) Bornm.	r	1.33
Asperula arvensis L.	r	0.77	*Nigella gallica* Jord.	r	0.61
Avena fatua L.	rrr	1.58	*Orlaya daucoides* (L.) Greut	rrr	-
Bifora radians M. Bieb.	rrr	0.15	*Papaver argemone* L.	rr	0.72
Bifora testiculata (L.) Spreng.	r	0.66	*Papaver dubium* L.	r	0.10
Biscutella auriculata L. subsp. *auriculata*	r	0.10	*Papver hybridum* L.	r	4.55
Bromus secalinus L.	r	0.05	*Polycnemum arvense* L.	rr	2.25
Bupleurum lancifolium Hornem	rr	-	*Polygonum aviculare* subsp. *bellardii* (All.) O. Bolòs et Vigo	rr	-
Bupleurum rotundifolium L.	rr	0.72	*Raphanus raphanistrum* L.	r	1.23
Camelina sativa subsp. *sativa* (L.) Crantz	r	-	*Roemeria hybrida* (L.) DC.	r	1.23
Centaurea collina L. subsp. *serratulifolia* (Sennen et Pau ex Hayek) Hayek	r	0.36	*Satureja rotundifolia* (Pers.) Briq.	rrr	-
Centaurea cyanus L.	r	1.12	*Silene conoidea* L.	rr	0.05
Cerastium perfoliatum L.	rr	-	*Silene muscipula* L.	r	0.05
Chrozophora tinctoria (L.) Raf.	rr	0.26	*Sinapis arvensis* L.	rr	0.46
Conringia orientalis (L.) Dumort	r	-	*Sisymbrium orientale* L. subsp. *orientale*	r	1.38
Delphinium peregrinum L.	r	0.61	*Thlaspi arvense* L. subsp. arvense	r	-
Delphinium pubescens DC.	r	0.51	*Turgenia latifolia* (L.) Hoffm.	rr	0.05
Filago arvensis L.	rr	-	*Vaccaria hyspanica* (Mill.) Rauschert	rr	4.14
Fumaria densiflora DC.	rr	0.51	*Valerianella coronata* (L.) DC.	rr	0.10
Fumaria vaillantii Loisel. in Desv.	rr	-	*Valerianella echinata* (L.) DC.	rrr	-
Gagea villosa (M. Bieb.) Duby	r	*	*Valerianella pumila* (L.) DC.	r	-
Galium aparine subsp. *spurium* (L.) Simonk	r	15.07	*Vicia benghalensis* (L.)	r	0.10
Gladiolus italicus Mill.	rr	0.46	*Vicia bithynica* (L.) L.	r	0.10
Glaucium corniculatum (L.) J.H. Rudolph	rr	0.20	*Vicia pannonica* Crantz subsp. s*triata* (M. Bieb.) Nyman	r	-
Iberis pinnata L.	rr	-	*Vicia villosa* subs. v*aria* (Host) Corb.	r	11.24
Hypecoum pendulum L.	rr	0.77			

* Blooms earlier than the sampling period

Fe d’errades

On consta *Papver hybridum* L. ha de constar *Papaver hybridum* L.).

Capítol 3

Herbicide impact on the growth and reproduction of characteristic and rare arable weeds of winter cereal fields

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RESUM

Impacte dels herbicides en el creixement i la reproducció d’espècies arvenses característiques i rares dels camps cerealistes de secà

L’aplicació d’herbicides ha estat considerada una de les pràctiques agrícoles que ha afectat més negativament la diversitat d’espècies arvenses, sobretot d’aquelles que són especialistes dels camps cerealistes de secà. Si bé l’efecte de la deriva d’herbicides sobre les espècies dels hàbitats adjacents ha estat àmpliament testat, són molt pocs els estudis que examinen els efectes dels herbicides sobre les espècies arvenses característiques dels camps de cereals que han esdevingut rares i que, per tant, requereixen una atenció especial pel que fa a la seva conservació. La toxicitat dels herbicides normalment és avaluada segons certs criteris a curt termini, sobretot la biomassa. Però la sensibilitat als herbicides a curt termini no és suficient per detectar els efectes reals, ja que les plantes poden o no recuperar-se al llarg del temps després de l’aplicació d’herbicides a dosis subletals. Per aquest motiu, convé avaluar els efectes de l’herbicida a més llarg termini. Fins i tot és important determinar els efectes que pot arribar a tenir l’herbicida sobre la reproducció, especialment en el cas d’espècies anuals que depenen de la producció de llavors per assegurar la seva persistència, com són la majoria d’espècies arvenses característiques dels cultius cerealistes de secà.

En aquest estudi es van avaluar els efectes de dos herbicides d’ús habitual sobre quatre parelles d’espècies segetals, una espècie més rara i una més comuna, de famílies diferents. L’elecció de parelles d’espècies facilita la interpretació de la comparació entre espècies rares i comunes, ja que aquest disseny té en compte que plantes filogenèticament properes poden respondre de manera similar davant els herbicides. Els nostres objectius van ser 1) examinar el patró de sensibilitat de les espècies seleccionades als herbicides testats a curt i a llarg termini segons la biomassa aèria total, la biomassa reproductiva i el nombre de llavors, 2) determinar els nivells i el temps de recuperació en termes de llargada de la tija i del nombre de fruits, de les plantes afectades per dosis creixents d’herbicides i 3) avaluar si les espècies arvenses rares són més sensibles a l’aplicació d’herbicides que les espècies relacionades més comunes.

Es va avaluar separadament els efectes de l’aplicació de dos herbicides molt utilitzats en els camps de cereals a Catalunya, el tribenuron i el 2,4-D, aplicats a sis dosis diferents, a part del controls. Es van preparar dos conjunts de plantes de cada condició per tal de fer l’avaluació a curt (28 dies després de l’aplicació d’herbicida) i a llarg termini (48 o 55 dies després de l’aplicació d’herbicida) de la biomassa i del nombre de llavors produïdes. A partir d’aquestes dades es van

estimar les concentracions d’inhibició per cada espècie mitjançant models logístics. Aquests models també es van ajustar per les dues espècies de la mateixa família juntes per avaluar les diferències entre les respostes als herbicides de l’espècie rara i la comuna. Es van prendre mesures de la llargada de la tija i del nombre de fruits produïts setmanalment durant l’experiment per avaluar la capacitat de recuperació. Mitjançant models lineals es van analitzar les diferències entre les mesures de les plantes tractades amb els herbicides i les control per cada setmana. Les plantes es van considerar recuperades quan, després de presentar diferències significatives, arribaven a ser iguals que els controls.

La majoria de les espècies testades va patir efectes significatius sobre la biomassa i la reproducció a dosis molt baixes d’ambdós herbicides. Atès que les espècies segetals habiten dins els camps, solen estar sotmeses a dosis molt més altes, el que probablement ha conduït a desplaçar-les cap a les vores dels camps, on les pràctiques agrícoles hi actuen de manera menys intensiva (Romero et al. 2008b). L’efecte dels herbicides, però, depèn de la dosi d’aplicació i de l’herbicida en qüestió, i també del moment d’avaluació, ja que per determinades espècies hem trobat certa capacitat de recuperació, com en altres estudis (Riemens et al. 2009; Carpenter et al. 2013). La capacitat de recuperació depèn tant de l’herbicida com de la dosi d’aplicació; el temps de recuperació és més llarg com més altes siguin les dosis d’herbicides. Tot i això, la capacitat de recuperació és molt variable entre espècies, sense una relació directa amb el seu grau de raresa. Els efectes dels herbicides en la producció de llavors en general van ser més remarcables que en la biomassa, com ja s’havia trobat prèviament per altres espècies (Boutin et al., 2014; Carpenter and Boutin, 2010; Clark et al., 2004; EFSA PPR Panel, 2014). La davallada en la producció de llavors d’aquestes espècies anuals pot tenir efectes sobre el seu banc de llavors, fet que comporta reduccions en el reclutament futur (EFSA PPR Panel, 2014).

Amb aquests resultats podem afirmar que l’aplicació d’herbicides ha estat un determinant important de la davallada de la diversitat vegetal als sistemes agrícoles. Malgrat tot, la raresa en si no presenta una relació directa amb la sensibilitat als herbicides. Per aquest motiu, convé cercar les causes de l’enrariment de les espècies arvenses característiques en la coincidència de diversos factors, d’entre els quals l’aplicació d’herbicides hi deu haver tingut un paper important (Royo-Esnal et al. 2011).

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Herbicide impact on the growth and reproduction of characteristic and rare arable weeds of winter cereal fields

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Abstract The decline of arable species characteristic of winter cereal fields has often been attributed to different factors related to agricultural intensification but most importantly to herbicide use. Herbicide phytotoxicity is most frequently assessed using short-term endpoints, primarily aboveground biomass. However, short-term sensitivity is usually not sufficient to detect actual effects because plants may or may not recover over time following sublethal herbicide exposures. Therefore, it is important to assess the long-term effects of herbicide applications. Annual species rely on renewable seed production to ensure their persistence; hence, assessment of herbicide sensitivity is more accurately estimated through effects on reproduction. Here we aim to assess the phytotoxicity of two commonly used herbicides: tribenuron and 2,4-D on eight plant species belonging to four families, each with one rare and one more common species. Specifically we examined the pattern of sensitivity using short-term and long-term endpoints (total aboveground biomass, total seed biomass and number of seeds) of these species; we determined the levels of and time to recovery in terms of stem length and fruit number, and assessed whether their rarity relates to their sensitivity to herbicide application. Our results suggest that although differences in herbicide sensitivity are not a direct cause of rarity for all species, it may be an important driver of declining arable plants.

Keywords Herbicide risk assessment · Plant recovery · Short- and long-term testing · Tribenuron · 2,4-D

Introduction

Weed and crop species compete for the same resources and, hence, tend to be exclusive. That is why most conventional agricultural practices, aimed at improving crop production, have led to remarkable reductions in arable plant populations and diversity (Freemark and Boutin 1995; Storkey et al. 2012). As a consequence, there are a number of species characteristic of arable land that, without being a real nuisance to crop production, have suffered important population declines to the point of becoming rare in arable fields (Baessler and Klotz 2006; Chamorro et al. 2007; Fried et al. 2009). The decline of arable species has often been attributed to different factors related to agricultural intensification (Hole et al. 2005; Romero et al. 2008; Storkey et al. 2012). Several studies have highlighted that amongst a variety of farming practices, herbicides often play a prominent role in the observed decrease of non-crop plant diversity in agricultural systems (e.g. Hyvönen and Salonen 2002). Plants outside crop fields can be affected by low dosages of herbicide, typically caused by herbicide drift and runoff from sprayed fields towards the adjacent non-target areas. The effect of herbicide misplacement on non-target plants has been reported in several studies (Marrs et al. 1991; de Snoo and van der Poll 1999; Riemens et al. 2008; Dalton and Boutin 2010; Schmitz et al. 2014a, b). Nevertheless, very few studies have

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examined the role of herbicide application on the species of arable fields that have recently become rare.

Effects of herbicide toxicity have often been assessed using short-term endpoints, usually aboveground biomass (Breeze et al. 1992; Carpenter and Boutin 2010; Egan et al. 2014), as promoted by the accepted guidelines (OECD 2006; USEPA 2012). Different studies, however, have demonstrated that short-term sensitivity is often not sufficient to detect actual effects because plants may or may not recover over time (Carpenter et al. 2013; Boutin et al. 2014). The recovery process following sublethal herbicide exposure can be slow and this could potentially reduce a plant's ability to compete with other more resistant or faster recovering species. Therefore, the latter species would likely be favored since they may be able to monopolize resources at the expense of those plants that are more affected (Boutin et al. 2010; Carpenter et al. 2013). The differences between species in the time required to recover might be indicative of the future species assemblages that arise following herbicide application.

Though some species are able to recover vegetatively at low exposure dosages, reproduction may continue to be affected, as it has been previously shown to be a more sensitive endpoint than vegetative parameters in some cases (Clark et al. 2004; Carpenter and Boutin 2010; Carpenter et al. 2013; Boutin et al. 2014). Reproductive parameters should especially be considered when testing herbicide phytotoxicity on annual species, because annuals rely heavily on reproduction to ensure future population recruitment opportunities (Carpenter et al. 2013; EFSA PPR Panel 2014). Herbicide application can thus lead to long-term consequences on the plant community of arable habitats, potentially by changing the amounts and types (i.e. species) of viable seeds that are found in the soil seedbank (Carpenter and Boutin 2010). As such, populations of highly susceptible species may be greatly reduced or replaced by the more tolerant ones (Freemark and Boutin 1995; Crone et al. 2009).

The conservation of rare arable species must be considered carefully in agricultural areas. They have high intrinsic values as components of biodiversity and as key agro-environmental indicators of arable cropping systems. In addition, because these rare species are more diverse than the few crop species that normally dominate arable land, they provide a greater variety of resources and functions in agroecosystems, such as a source of pollen to attract more pollinators, or plant biomass or fruits for herbivores, predators and parasitoids (Hawes et al. 2003; Tscharntke et al. 2005).

Here we aim to study the effects of herbicides on the conservation of rare arable species by assessing the phytotoxicity of two commonly used herbicides in Mediterranean arable cereal crops (tribenuron and 2,4-D) on selected arable species. The eight species tested belong to four plant families, from which we selected one rare and one common species. This test allowed us to take into account the fact that close plant relatives may respond similarly to herbicides, thus making fairer the comparison between the paired common and rare species (Clark et al. 2004). Specifically, our objectives were (1) to examine the pattern of sensitivity of short-term and long-term endpoints (total aboveground biomass, seed biomass and number of seeds) of selected arable weed species; (2) to determine the levels of and time to recovery, in terms of both stem length and fruit number, for plants affected by increasing doses of each herbicide and (3) to assess whether the sensitivity to herbicide application is higher in rare characteristic arable weeds than in more common related species.

Materials and methods

Experimental set up

All experimental work was conducted in the greenhouses of the National Wildlife Research Centre (Environment Canada), Ottawa, ON, between February and May 2012 and between April and June 2013. Average daily temperature ranged from 17.2 ± 0.3 to 34.2 ± 1.1 °C and the photosynthetic active radiation (PAR) ranged from 285 (cloudy day) to 1951 μmol photons m^{-2} s^{-1} (sunny day).

Four pairs, consisting each of one rare and one common plant species, of phylogenetically close arable weed species were tested in this experiment (Table 1). Species selected were considered either common or rare, according to field frequency records in an extensive survey of arable fields in north-eastern Spain (Rotchés-Ribalta et al. 2014). Seeds were collected in the spring-summer of 2011 from plant populations found in different cereal fields located within the north-eastern Iberian Peninsula. Seeds of all species were surface sown separately in small trays containing a 3:1 (by volume) soil mixture consisting of Pro-Mix® BX (Premier Horticulture Ltd., Rivière-du-Loup, Québec, Canada) potting soil and silica sand (Opta Minerals Inc., Waterdown, Ontario, Canada). Trays of seeds requiring stratification (*Asperula arvensis*, *Papaver rhoeas*, *Papaver argemone*, *Bupleurum rotundifolium* and *Scandix pecten-veneris* subsp. *pecten-veneris* (hereinafter *S. pecten-veneris*)) were first placed into a dark 2–4 °C refrigerator for 10 days prior to being moved into a growth chamber set at a 10 h daylight:14 h night cycle at temperatures varying from 20 to 10 °C, respectively, to promote germination. Once high germination rates were obtained seedlings (with cotyledons) were transplanted singly into 10 cm diameter by 9 cm high pots ensuring that enough healthy plants were available for all experimental conditions. Pots contained

Table 1 Pairs of common and rare arable weed species found in arable fields in north-eastern Spain used in this study and their frequency of occurrence according to their presence in 1957 sampled plots in a previous study (Rotchés-Ribalta et al. 2014)

Family	Species	Frequency (%)	Rarity
Apiaceae	*Bupleurum rotundifolium* L.	0.72	Rare
	Scandix pecten-veneris L. subsp. *pecten-veneris*	15.79	Common
Brassicaceae	*Neslia paniculata* (L.) Desv. subsp. *thracica* (Velen.) Bornm.	1.33	Rare
	Rapistrum rugosum (L.) All.	8.02	Common
Papaveraceae	*Papaver argemone* L.	0.72	Rare
	Papaver rhoeas L.	61.01	Common
Rubiaceae	*Asperula arvensis* L.	0.77	Rare
	Galium aparine L. subsp. *spurium* (L.) Simonk	15.07	Common

the same 3:1 soil and silica sand mixture as for germination.

The experimental setup consisted of two herbicides (tribenuron and 2,4-D) tested separately at six different doses plus a set of controls. For each herbicide dose, five plant replicates each consisting of one plant per pot were used. For each species, half the plants was used to address short-term effects, while the other half was used to address long-term effects (i.e. two harvest times). A total of 140 seedlings were used for each species. Overall, eight plant species were tested, which accounted for a total of 1120 plants in the experiment.

Herbicide information and application

We tested the phytotoxicity of Spartan® herbicide, containing tribenuron methyl (methyl 2-[[[[(4-methyl-1,3,5-triazin-2-yl)methylamino]carbonyl]amino]sulfonyl]benzoate, CAS number 101200-48-0) at 0.75 g acid-equivalent (a.e.) L^{-1} (E.I Dupont Canada Company, Mississauga, Ontario, Canada) and 2,4-D amine 600 (2,4-Dichlorophenoxyacetic acid, dimethylamine salt, with CAS number 2008-39-1) liquid herbicide composed of dimethylamine salt at 564 g a.e. L^{-1} (Nufarm Agriculture Inc., Calgary, Alberta, Canada). Tribenuron is a selective post-emergence herbicide used to control broad-leaf plants in cereal fields. It is a group 2 herbicide acting by inhibiting acetolactate synthase (ALS), a key enzyme in the biosynthesis of the branched-chain amino acids (Larossa and Schloss 1984) thus stopping cell growth and division. 2,4-D is a selective post-emergence herbicide used to control broad-leaf plants in cereal and sugarcane fields and grass pastures. 2,4-D is a group 4 herbicide mimicking the endogenous auxin (IAA). Its mechanism of action is not well known but it appears to affect both cell wall plasticity by acidification and nucleic acid metabolism by stimulating RNA polymerase (WSSA 2014).

The herbicide application rates were 7.5 g a.e. ha^{-1} for tribenuron and 564 g a.e. ha^{-1} for 2,4-D, based on recommendations provided on the label (Du Pont Canada Company and NuFarm Agriculture Inc. respectively). Six 500 mL solutions of each herbicide were prepared following a geometric progression of two for both herbicides. For tribenuron we prepared herbicide solutions with: 0.234, 0.469, 0.938, 1.875, 3.75 and 7.5 g a.e. ha^{-1} and for 2,4-D: 17.625, 35.25, 70.5, 141, 282 and 564 g a.e. ha^{-1}. A nonionic surfactant, Agral® 90 (Norac Concepts) containing nonylphenoxy polyethoxyethanol was added to tribenuron methyl, as recommended on the label, to improve the coverage of herbicide spray mixtures. No surfactant was used with 2,4-D since it is not recommended on the label. Once made, all herbicide solutions were used within a week, after which fresh batches were prepared.

Herbicides were applied using a track spray-booth (de Vries Manufacturing, Hollandale, MN, USA) equipped with a TeeJet 8002E flat-fan spray nozzle (Spraying Systems, Wheaton, IL, USA). The system was calibrated prior to herbicide application to ensure that 6.75 mL m^{-2} of solution was delivered at a pressure of 206.84 kPa. Plants were sprayed when seedlings had reached the four to six true leaf stage. Prior to spraying, plants were sorted and grouped by size across all doses and harvest times (short-term and long-term plants) to ensure size uniformity. Plants were then randomly assigned numerical ID tags to prevent potential bias during measurements. All plants were well hydrated prior to spraying in order to maintain humidity for herbicide absorption. Both short- and long-term plants were sprayed at the same time using the same prepared herbicide solutions. The control plants were not sprayed but well hydrated. Newly sprayed plants were kept isolated from the main experimental greenhouses to avoid potential effects of volatilization and drift of the herbicides and were not watered for a 24 h period to optimize herbicide absorption. All plants of a given species were randomized on a weekly basis by dose/treatment within blocks in the greenhouse to ensure uniformity of growing conditions.

Assessing short-term effects on plant biomass

Short-term assessment of aboveground biomass is often the most common and preferred endpoint used in phytotoxicity testing because current guidelines rely heavily on short-term evaluations (OECD 2006; USEPA 2012). To assess short-term effects of tribenuron and 2,4-D on plant biomass, all plants belonging to the short-term treatment were harvested 28 days after herbicide spray. Plants were cut at the soil level and necrotic tissues resulting from herbicide exposure were removed so as not to be confused with live plant biomass without herbicide affectation. All aboveground plant material obtained was bagged separately and dried in a drying oven at approximately 70 °C for at least 3 days before being weighed.

Assessing long-term effects on plant biomass

The long-term harvest was carried out for a given species when the plants had finished reproducing and the controls began to show signs of natural senescence. This corresponded to 48 days after herbicide application, with the exceptions of *B. rotundifolium* and *S. pecten-veneris*, which began to senesce 55 days after exposure. All aboveground plant material, except seeds, was bagged, dried and weighed, following the same protocol as for the short-term harvest. Before drying, all the seeds of each plant were counted and bagged separately. Some seeds (50 or 100) of each individual were weighed for fresh weight and then dried in the oven at 70 °C for at least 3 days to obtain the dry weight. The fresh/dry weight ratio measure was used to extrapolate dry weights of the rest of the seeds to get the total seed biomass. In the case of *P. rhoeas* and *P. argemone* we also used this measure to extrapolate the total number of seeds. Dry weights of all aboveground tissues, including vegetative and reproductive parts were summed to obtain total long-term aboveground biomass.

Assessing recovery

During the course of the experiment weekly measurements of stem length, number of fruits (for the Papaveraceae, Brassicaceae and Apiaceae) and whorls of fruits (for *A. arvensis* L.) were taken to assess plant recovery. Stem length measures were taken from the fully extended main stem of the plants. Measurements of stem length started at week 0 for *Galium aparine* subsp. *spurium* (hereinafter *G. spurium*) and *A. arvensis*; for the six remaining species, stem length was determined later during the natural life cycle after bolting had occurred. The number of fruits was collected over time for all species except *G. spurium*, for which all fruit were collected and counted on the final day of the long-term treatment. Due to the lack of pollinators within the greenhouses, fruit and seed production of *P. rhoeas* and to a lesser extent *Rapistrum rugosum* was achieved by artificial pollination using a small paint brush.

Statistical analysis

Inhibition concentrations

We described the response of sprayed plants to herbicide dosage by means of logistic models (Environment Canada 2005) described in Annex 1. We assumed that the maximum response, corresponding to the upper asymptote of the model (*Asym*), was achieved for the unsprayed controls and, thus, we assigned the mean of biomass, total seed biomass or number of seeds of the unsprayed controls to this parameter. Inhibition concentration values IC50, IC25 and IC10, defined as the dosage that causes a 50, 25 and 10 % reduction in growth (biomass) or reproduction (seed number or seed biomass), respectively, of a plant as compared to unsprayed controls, were fitted through nonlinear regression.

The models were fitted for all species and treatments to the total biomass and to seed number and biomass using R 3.0.3 (R Core Team 2013) with the package nlme (Pinheiro et al. 2013) for non-linear mixed-effects models. Data was square-root transformed to ensure normality and homoscedasticity of the residuals. No IC values were obtained when the data did not fit to the regression logistic model.

Comparison between rare and common species

To evaluate potential differences between rare and common species of a given family, we fitted logistic models to the dose–response of growth and reproductive parameters (biomass, total seed biomass and number of seeds) for each pair of species. We determined whether there were significant differences in herbicide sensitivity as measured by the location parameter IC25 (xmid) and the slope or scale parameter (scal) between the common and the rare species tested. The models were fitted to the rare and the common species of each family jointly, including two extra parameters which quantify the difference between species in IC25 and scale parameters. Thus, the fitted model provides a statistical test for these differences between species. We also used the R 3.0.3 program (R Core Team 2013) with the package nlme (Pinheiro et al. 2013).

Evaluation of time to recovery

Linear models were used to analyze the differences in stem length and fruit number between treated plants and untreated controls for each week. Therefore, we determined the length of time required for vegetative (stem length over time) or reproductive parameters (fruit number or whorls of fruits over time) to recover. Plants were considered recovered when the affected plants no longer significantly differ from the controls. For all analyses, the model assumptions of normality of residuals and homogeneity of variance were verified. These analyses were carried out in R 3.0.3 (R Core Team 2013).

Results

Herbicide sensitivity

Herbicide sensitivity was assessed as IC25 (Tables 2, 3). IC50 and IC10 results are presented in annexes 2 and 3. The aboveground biomass at the short-term harvest of six of the eight species treated with tribenuron suffered a 25 % reduction at doses ranging from 1.9 to 43.5 % of the recommended label rate (Table 2). The plants that were not affected by tribenuron were the two rare species *Neslia paniculata* subsp. *thracica* (hereinafter *N. paniculata*) (Brassicaceae) and *A. arvensis* (Rubiaceae). In the long-term harvest, however, effects were generally reduced, such that after 48 (or 55) days after herbicide application only the Papaveraceae and the Apiaceae species were still noticeably impacted. Both *R. rugosum* and *G. spurium*, that experienced significant 25 % reductions in biomass in the short-term, were able to recover biomass in the long-term.

The herbicide 2,4-D caused a negative effect on five of the eight species tested in terms of both the short- and long-term aboveground biomass parameter (Table 2). The Rubiaceae species were not affected by 2,4-D and neither was *P. rhoeas*. The majority of affected species had IC25 values below 50 % of the recommended label rate in both the short- and long-term.

Significant differences were found between common and rare species of the same family. However, the direction of the effect was not as expected in all the cases (Fig. 1). Tribenuron did not affect the rare *N. paniculata* and *A. arvensis* species both in the short- and in the long-term. There were significant differences with their respective common species *R. rugosum* and *G. spurium* in the short-term, however, this disappeared in the long-term. On the contrary, tribenuron caused a significant decrease in the biomass of the Apiaceae and Papaveraceae (both the common and the rare), whereby in both cases the rare species was significantly more affected. 2,4-D caused remarkable effects on both common and rare species of the Apiaceae, Brassicaceae and Papaveraceae. But it was only the rare *P. argemone* which was significantly more affected by this herbicide than its paired common species. For the Rubiaceae, the common *G. spurium* was significantly more affected than *A. arvensis* in the short-term when 2,4-D was applied, but it was able to recover over time, such that no statistical differences were found between them in the long-term.

Effect on reproduction

Generally, seed biomass and seed number were reduced by 25 % at lower doses of herbicides than the vegetative biomass variable, except for *B. rotundifolium* (Tables 2, 3). Only *N. paniculata* and *A. arvensis* had produced seeds at the time of the short-term harvest (28 days after the herbicide spray). The short-term seed production of these two species was not affected by tribenuron dosage; however, high doses of 2,4-D did cause marked reductions in the seed production and reproductive biomass of *N. paniculata*.

In the long-term, both *S. pecten-veneris* and *B. rotundifolium* seed production were affected by the herbicides tested. *B. rotundifolium* was more affected than *S. pecten-veneris* under tribenuron, but no statistically significant differences were found for 2,4-D (Fig. 2). *Neslia paniculata* total seed biomass in the long-term harvest showed no effect of tribenuron but it was affected by 2,4-D. However, we were not able to compare reproduction of *N. paniculata* to the common *R. rugosum* since the last one failed to produce sufficient amounts of seed. Both *Papaver* species showed strong reductions in seed production with increasing dosages of both herbicides. There was no detectable difference between *P. rhoeas* and *P. argemone* responses in the tribenuron treatments but there were statistically significant differences between them for 2,4-D, with the rarer *P. argemone* showing a greater negative effect on total seed biomass. High doses of tribenuron caused a reduction in seed biomass of *G. spurium*, but not of *A. arvensis*, revealing significant differences between these two species. On the other hand, 2,4-D showed no noticeable effect on the reproduction of either of these Rubiaceae species (Fig. 2).

Recovery

The weekly stem length measures allowed for an assessment of the vegetative recovery over time for all species. Similarly, weekly fruit (or whorls of fruits for *A. arvensis*) counts allowed for an assessment of reproductive recovery over time following herbicide exposure. *Papaver* species were more affected by tribenuron than by 2,4-D herbicide to the extent that they were unable to recover, both in terms of stem length and fruit production, from tribenuron application during the eight week span of the experiment (Tables 4, 5). *Papaver argemone* was more affected by 2,4-D than *P. rhoeas*, and was unable to recover at doses higher than 25 % of the label rate. *B. roundifolium* did not

Table 2 Summary of the IC25s (defined as the dosage that resulted in a 25 % reduction in the overall aboveground biomass) for species tested with tribenuron and 2,4-D

	Species	Test duration	IC25 (g a.e. ha^{-1})	95 % confidence intervals	% of label rate
Tribenuron					
Apiaceae	*Bupleurum rotundifolium* [R]	ST	0.986	0.709–1.307	13.142
		LT	1.107	0.739–1.553	14.763
	Scandix pecten-veneris subsp. *pecten-veneris* [C]	ST	2.449	1.656–3.479	32.654
		LT	5.575	3.793–8.021	74.334
Brassicaceae	*Neslia paniculata* subsp. *thracica* [R]	ST	NE		
		LT	NE		
	Rapistrum rugosum [C]	ST	0.377	0.251–0.517	5.033
		LT	NE		
Papaveraceae	*Papaver argemone* [R]	ST	0.144	0.110–0.179	1.922
		LT	0.317	0.245–0.392	4.221
	Papaver rhoeas [C]	ST	0.404	0.196–0.649	5.388
		LT	0.598	−0.627–5.854	7.976
Rubiaceae	*Asperula arvensis* [R]	ST	NE		
		LT	NE		
	Galium aparine subsp. *spurium* [C]	ST	3.260	1.807–5.467	43.473
		LT	>7.500		
2,4-D					
Apiaceae	*Bupleurum rotundifolium* [R]	ST	273.429	200.351–373.029	48.480
		LT	285.200	211.371–384.696	50.567
	Scandix pecten-veneris subsp. *pecten-veneris* [C]	ST	171.744	126.565–232.925	30.451
		LT	142.473	100.838–201.131	25.261
Brassicaceae	*Neslia paniculata* subsp. *thracica* [R]	ST	151.240	118.438–193.052	26.816
		LT	192.508	155.736–237.909	34.133
	Rapistrum rugosum [C]	ST	76.334	62.029–93.885	13.534
		LT	137.293	82.799–227.223	24.343
Papaveraceae	*Papaver argemone* [R]	ST	347.859	265.380–455.877	61.677
		LT	173.181	82.732–361.333	30.706
	Papaver rhoeas [C]	ST	>564.000		
		LT	NE		
Rubiaceae	*Asperula arvensis* [R]	ST	NE		
		LT	NE		
	Galium aparine subsp. *spurium* [C]	ST	>564.000		
		LT	NE		

The label rate for tribenuron was 7.5 g acid-equivalent (a.e.) ha^{-1} and for 2,4-D was 564 g a.e. ha^{-1}. 'ST' indicates the short-term and 'LT' the long-term harvest data. We used '>7.500' and '>564.000' when the estimated IC25s were greater than the highest dose evaluated for each herbicide. 'NE' indicates no statistically significant effect. The rarity of each species is indicated as [R] rare or [C] common

recover in either stem length or fruit production at tribenuron application rates of 6.25 % label rate and higher; conversely, for 2,4-D, stem length only failed to recover at 100 % dose, while reproduction was never greatly affected. The common *S. pecten-veneris*, in contrast, was able to recover vegetatively at all doses, slowly over time (seven to nine weeks) following the tribenuron treatment, but neither its stem length nor reproduction did recover at higher doses of 2,4-D (Tables 4, 5). The rare species *A. arvensis* experienced no negative effects on either stem length or fruit production following both herbicides application (Tables 4, 5). On the contrary, the common species *G. spurium*, although more negatively affected than *A. arvensis*, was able to recover in terms of stem length

Table 3 Summary of the IC25s (defined as the dosage that resulted in a 25 % reduction in total seed biomass and number of seeds) for species tested with tribenuron and 2,4-D

	Species	Test duration	Variable	IC25 (g a.e. ha^{-1})	95 % confidence intervals	% of label rate
Tribenuron						
Apiaceae	*Bupleurum rotundifolium* [R]	LT	Seed biomass	0.099	−0.329–0.798	1.314
			Num of seeds	NE		
	Scandix pecten-veneris subsp. *pecten-veneris* [C]	LT	Seed biomass	1.837	1.092–2.846	24.487
			Num of seeds	NE		
Brassicaceae	*Neslia paniculata* subsp. *thracica* [R]	ST	Seed biomass	NE		
			Num of seeds	NE		
		LT	Seed biomass	NE		
			Num of seeds	NE		
Papaveraceae	*Papaver argemone* [R]	LT	Seed biomass	0.227	0.209–0.245	3.024
	Papaver rhoeas [C]	LT	Seed biomass	0.126	−0.054–0.340	1.682
Rubiaceae	*Asperula arvensis* [R]	ST	Seed biomass	NE		
			Num of seeds	NE		
		LT	Seed biomass	NE		
			Num of seeds	NE		
	Galium aparine subsp. *spurium* [C]	LT	Seed biomass	6.690	3.742–11.469	89.194
			Num of seeds	NE		
2,4-D						
Apiaceae	*Bupleurum rotundifolium* [R]	LT	Seed biomass	>564.000		
			Num of seeds	NE		
	Scandix pecten-veneris subsp. *pecten-veneris* [C]	LT	Seed biomass	81.433	31.652–207.110	14.438
			Num of seeds	NE		
Brassicaceae	*Neslia paniculata* subsp. *thracica* [R]	ST	Seed biomass	98.885	71.631–136.365	17.533
			Num of seeds	65.298	39.047–108.757	11.578
		LT	Seed biomass	182.569	120.090–277.287	32.370
			Num of seeds	169.059	91.553–311.470	29.975
Papaveraceae	*Papaver argemone* [R]	LT	Seed biomass	25.934	5.896–104.202	4.598
			Num of seeds	12.338	1.671–65.596	2.188
	Papaver rhoeas [C]	LT	Seed biomass	NE		
			Num of seeds	NE		
Rubiaceae	*Asperula arvensis* [R]	ST	Seed biomass	>564.000		
			Num of seeds	NE		
		LT	Seed biomass	>564.000		
			Num of seeds	NE		
	Galium aparine subsp. *spurium* [C]	LT	Seed biomass	>564.000		
			Num of seeds	NE		

The selected label rate for tribenuron was 7.5 g acid-equivalent (a.e.) ha^{-1} and for 2,4-D was 564 g a.e. ha^{-1}. 'ST' indicates the short-term and 'LT' the long-term harvest data. We used '>7.500' and '>564.000' when the estimated IC25s were greater than the highest dose evaluated for each herbicide. 'NE' indicates no statistically significant effect. The rarity of each species is indicated as [R] rare or [C] common

within 2–6 weeks after exposure, though tending to require increasingly longer periods of recovery at higher dosages. Brassicaceae species exhibited similar responses as the Rubiaceae, except that at higher doses of both herbicides *R. rugosum* plants failed to recover during the course of their life cycle (Tables 4, 5).

Discussion

Herbicide application is one of the leading causes of the declining weed species diversity and abundance observed in agricultural landscapes, as well as a factor exerting changes in the composition of weed communities

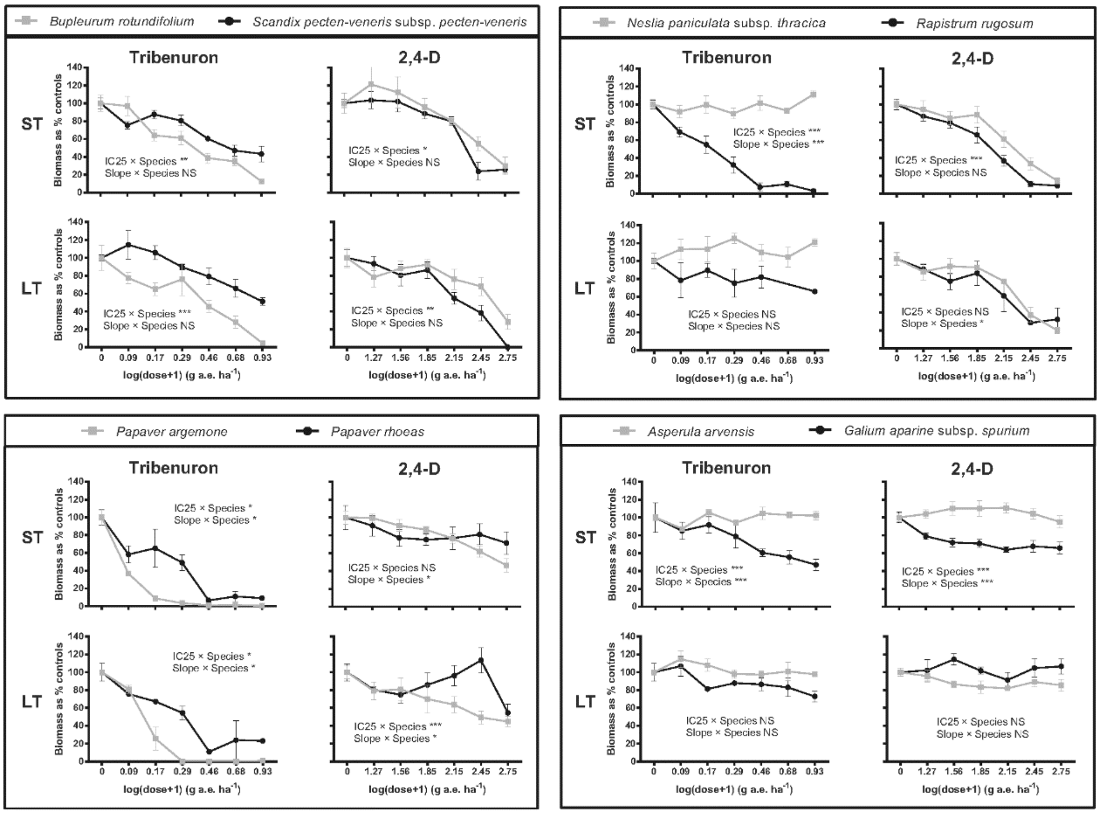

Fig. 1 Dose-response curves for species tested with the herbicides tribenuron and 2,4-D grouped by plant family: the rare species are represented in *grey* and the common species in *black*. *Curves* represent the percent average biomass of the sprayed plants as compared to the untreated controls. For each species pair, *top row graphs* represent the short-term biomass (ST) while those on the *bottom* depict the long-term biomass (LT). Significance of the effect of the dose is indicated as * when p-value <0.05; **<0.01; ***<0.001 and 'NS' when no significance was found. The interactions between the species and the IC25 (inhibition concentration of herbicide causing a 25 % biomass reduction) and the slope of the dose–response curve were used to assess differences between the paired species

(Hyvönen and Salonen 2002; José-María et al. 2011). Most species tested in this experiment suffered 25 % damage to biomass or reproduction at very low doses of both herbicides, often at doses below 50 % of recommended label rates. Considering that arable weed species thrive preferentially in crop fields, their populations probably have to withstand recurrent high doses of herbicides, especially in areas where multiple herbicide applications are performed each year. It has been suggested that the most sensitive species, the ones that are most affected by herbicide applications, may have been displaced at the expense of the more tolerant species (Freemark and Boutin 1995; Crone et al. 2009). In fact, it is likely that most herbicide sensitive species are now only found at the edges of organic fields, where agricultural practices are performed less intensively (Romero et al. 2008).

Our data showed that, as expected, the sensitivity of arable weed species to herbicides depended both on the dose of application and on the type of herbicide applied (Clark et al. 2004; Carpenter and Boutin 2010). Applications of tribenuron and 2,4-D at the juvenile stage of development caused significant reductions in total aboveground biomass in the majority of the species tested when measured after 4 weeks following herbicide application (short-term). These short-term effects have been documented in a large number of species following exposure to different herbicides (Breeze et al. 1992; Boutin et al. 2010; Egan et al. 2014). Effects of tribenuron on the aboveground biomass of sprayed plants 8 or 9 weeks after initial exposure (long-term effect) were less noticeable than those in the short-term. This indicates that some of the species tested (i.e. *G. spurium* and *R. rugosum*) were able

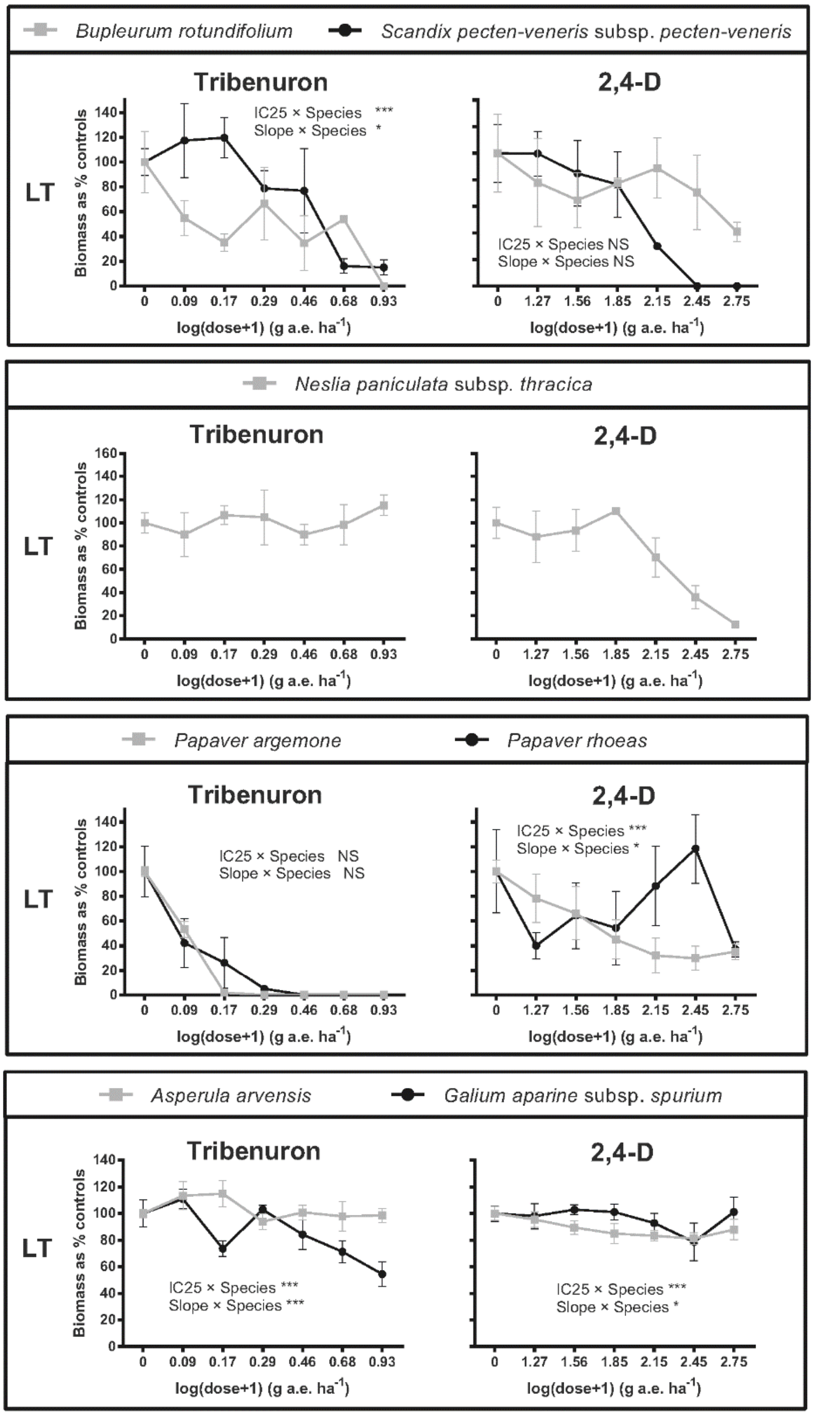

Fig. 2 Dose-response curves for species tested with the herbicides tribenuron and 2,4-D grouped by plant family: the rare species are represented in *grey* and the common species in *black*. *Curves* represent the percent average total seed biomass of the sprayed plants as compared to the untreated controls in the long-term harvest (LT). Significance of the effect of the dose is indicated as * when p-value < 0.05; ** < 0.01; *** < 0.001 and 'NS' when no significance was found. The interactions between the species and IC25 (inhibition concentration of herbicide causing a 25 % biomass reduction) and the slope of the dose–response curve were used to assess differences between the paired species

to recover their biomass in at least some tribenuron doses after initially being damaged, as has also found for other species and herbicides (Riemens et al. 2009; Carpenter et al. 2013). This recovery of biomass, however, was less pronounced when 2,4-D was applied.

Considering that herbicides have been listed as one of the main causes of the loss of arable weeds, we expected that the species that have undergone more noticeable declines and that are currently considered to be rarer would have higher sensitivities to herbicide application at non-lethal doses than the more common species. However, contrary to our assumptions, overall, rarity itself had no direct link to herbicide sensitivity in our pairs of species. We found significant differences between rare and common species in their sensitivities to the two tested herbicides. However, these differences did not always follow the expected direction. Comparable results were found by Egan et al. (2014), who observed that many common species were more susceptible to herbicides than the rare ones while in the case of other pairs, the rare species were more affected. For instance, in our experiments, the rare species *P. argemone* and *B. rotundifolium* were more sensitive to tribenuron application than their paired common species, which would indicate that they would be more at risk when tribenuron is applied. On the other hand, the reverse trend was observed for the rare species *A. arvensis* and *N. paniculata* that were more tolerant to tribenuron than the common species. In the case of the Brassicaceae and Apiaceae families, both rare and common species were similarly affected by 2,4-D application. Thus, even if herbicides have been important drivers of plant diversity loss, they are unlikely to be the sole determinant in explaining the overall decline of the rare weed species (Royo-Esnal et al. 2011). The absence of a uniform response pattern in the sensitivity to herbicides tested among common and rare arable weeds suggests that other factors are contributing to the dramatic reduction in their populations.

A plant that receives a sublethal herbicide dosage may experience delays in flowering or fruiting, which could potentially reduce seed production and, in turn, impact the seedbank replenishment (Strandberg et al. 2012). In other cases, flowering could be advanced by herbicide reaching plants (Qian et al. 2014), as observed for other stress

Table 4 Time to recovery (weeks) of stem length as determined through linear models analyses of weekly stem length data

Tribenuron	Week measurements began	Weeks until recovered					
		Dose (g a.e. ha^{-1}) and [% of label rate]					
		0.234 [3.125]	0.469 [6.25]	0.938 [12.5]	1.875 [25]	3.75 [50]	7.5 [100]
Bupleurum rotundifolium [R]	0	7	+	+	+	+	+
Scandix pecten-veneris subsp. *pecten-veneris* [C]	6	NE	NE	NE	7	8	9
Neslia paniculata subsp. *thracica* [R]	2	4	NE	NE	NE	3	NE
Rapistrum rugosum [C]	4	7	+	+	+	+	+
Papaver argemone [R]	3	+	+	+	+	+	+
Papaver rhoeas [C]	3	7	8	+	+	+	+
Asperula arvensis [R]	0	NE	NE	NE	NE	NE	NE
Galium aparine subsp. *spurium* [C]	0	2	2	5	6	5	6
2,4-D	Week measurements began	17.625 [3.125]	35.25 [6.25]	70.5 [12.5]	141 [25]	282 [50]	564 [100]
Bupleurum rotundifolium [R]	0	NE	NE	NE	NE	2	+
Scandix pecten-veneris subsp. *pecten-veneris* [C]	6	NE	NE	NE	+	+	+
Neslia paniculata subsp. *thracica* [R]	2	NE	NE	NE	NE	+	+
Rapistrum rugosum [C]	4	7	8	7	+	+	+
Papaver argemone [R]	3	NE	NE	8	+	+	+
Papaver rhoeas [C]	3	NE	NE	NE	NE	NE	5
Asperula arvensis [R]	0	NE	NE	NE	NE	NE	8
Galium aparine subsp. *spurium* [C]	0	NE	4	5	+	5	5

Numbers indicate the minimum time (weeks after spray) at which the affected plants no longer significantly differ from the controls. '+' indicates that stem length did not recover during the experiment at a given dose; 'NE' indicates no statistically significant effect. The rarity of each species is indicated as [R] rare or [C] common

Table 5 Time to recovery (weeks) of plant reproduction (number of fruits or whorls of fruits for *A. arvensis*) as determined through linear models analyses of weekly seed data

Tribenuron	Week measurements began	Weeks until recovered					
		Dose (g a.e. ha^{-1}) and [% of label rate]					
		0.234 [3.125]	0.469 [6.25]	0.938 [12.5]	1.875 [25]	3.75 [50]	7.5 [100]
Bupleurum rotundifolium [R]	7	9	+	NE	+	+	+
Scandix pecten-veneris subsp. *pecten-veneris* [C]	7	NE	NE	NE	NE	NE	9
Neslia paniculata subsp. *thracica* [R]	3	NE	NE	NE	NE	NE	NE
Papaver argemone [R]	4	8	+	+	+	+	+
Papaver rhoeas [C]	5	+	+	+	+	+	+
Asperula arvensis [R]	4	NE	NE	NE	NE	NE	NE
Galium aparine subsp. *spurium* [C]	8	NE	NE	NE	NE	NE	NE
2,4-D	Week measurements began	17.625 [3.125]	35.25 [6.25]	70.5 [12.5]	141 [25]	282 [50]	564 [100]
Bupleurum rotundifolium [R]	7	NE	NE	NE	NE	NE	NE
Scandix pecten-veneris subsp. *pecten-veneris* [C]	7	NE	NE	NE	+	+	+
Neslia paniculata subsp. *thracica* [R]	3	NE	NE	NE	5	6	6
Papaver argemone [R]	4	NE	NE	6	6	+	+
Papaver rhoeas [C]	5	NE	NE	NE	NE	NE	+
Asperula arvensis [R]	4	NE	NE	NE	NE	NE	NE
Galium aparine subsp. *spurium* [C]	8	NE	NE	NE	NE	NE	NE

Numbers indicate the minimum time (weeks after spray) at which the affected plants no longer significantly differ from the controls. '+' indicates that fruit production did not recover during the experiment at a given dose; 'NE' indicates no statistically significant effect. The rarity of each species is indicated as [R] rare or [C] common

factors such as competition or drought for instance (Grime 1979). Our data on seed production, when available, generally denoted higher damage than total biomass, as also recorded in other studies (Clark et al. 2004; Carpenter and Boutin 2010; Boutin et al. 2014; EFSA PPR Panel 2014). The fact that some reproductive endpoints were more sensitive than biomass endpoints indicated that for some species, risk assessment based solely on aboveground biomass may underestimate true herbicide sensitivities. Given that the tested species are annual plants that rely on reproduction to ensure their survival, seed loss would directly relate to declines in seedbank size and subsequently affect future seedling recruitment (EFSA PPR Panel 2014). This could be the case for the rare species *B. rotundifolium* and *P. argemone* that showed high sensitivities on reproductive measures in relation to tribenuron. Thus, herbicide application may have compromised the permanence of their populations and, by extension, of the species (Carpenter and Boutin 2010). The more common species *P. rhoeas*, nevertheless, was affected by tribenuron in the same way as *P. argemone*; however, field populations of *P. rhoeas* have not undergone severe reductions. *Papaver rhoeas* seed production was not affected by the application of 2,4-D, unlike *P. argemone*, which was affected and unable to recover seed production in the course of the experiment.

The patterns in recovery assessed through stem length and the number of fruits produced over time were similar for both variables. Stem length recovery was dependent on the herbicide dose applied, as expected, with plants at higher doses requiring more weeks to recover. Nevertheless, the recovery capacity and the time required to recover was highly variable amongst species, with no obvious relation to the rarity of the species. Similar results were found in Follak and Hurle (2004) and Carpenter et al. (2013). From a biological standpoint, sensitivity to herbicides and the ability to recover after damage must be understood within a community context to fully understand how populations of plants may be affected (Dalton and Boutin 2010). In natural communities, interspecific competition for light and space would, in theory, favor the more herbicide resistant species or those species that are faster to recover following sublethal exposures (Carpenter and Boutin 2010; Carpenter et al. 2013; Egan et al. 2014). Although our tests did not take into account direct competition between species, since plants were grown individually, we can infer how these species could potentially react in a community context. For instance, *P. rhoeas* plants would be at an advantage with respect to the rarer *P. argemone* when 2,4-D is applied, which highlights the relevance of this herbicide in affecting the rarity of this species. However, the common species *R. rugosum* would be more affected than its closely related rare species in a community where these herbicides are applied. Given these differences, to fully address the links between herbicide application and arable weed rarity, community level experiments should be conducted.

These findings must be interpreted cautiously for several reasons. Our experiments only tested eight species in four plant families, and therefore they represent just a small sample of all the species that we consider characteristic of arable fields (Rotchés-Ribalta et al. 2014). Moreover, we only tested two commonly used herbicides from a long list of herbicides that are frequently applied in the study area (Taberner 2013). Thus, though a species may be somewhat tolerant to one herbicide, all negative effects from multiple herbicides could be causing the actual decline. In addition, herbicides were sprayed at the juvenile stage of plant growth; past studies have demonstrated that herbicides reaching plants at the flower bud stage may be more detrimental in terms of plant reproduction (Fletcher et al. 1996; Boutin et al. 2014). Further phytotoxicity studies on rare arable plant species should therefore consider more species from different families conducted with different herbicides typically applied in arable crops, at various phenological stages and tested in community experiments.

Conclusions

Our results suggest that herbicides may be a prominent factor contributing to arable plant diversity decline since most of our study species suffered negative effects following herbicide application. However, other factors may have been determinant in reducing the populations of some arable weeds that have become rare. The interaction of the different agricultural management practices, such as fertilizer application and mechanization, may have also played a significant role (Storkey et al. 2012). Giving that the rare species showed different responses to the application of two commonly used herbicides, we can postulate that the rarity of each of these species has been driven by a variety of idiosyncratic factors.

Since arable weed species show very specific responses to agricultural management practices, future research should consider herbicide phytotoxicity on these arable species to ascertain the effects of herbicide exposure on their populations. Although we found limited linkage between herbicide application and the rarity of arable weeds studied in this experiment, herbicides remain an important driver of declining arable plant diversity. Therefore, more trials should be conducted to test the tolerance of these arable species to the different herbicides that are regularly applied in fields to ascertain how they respond to the applications. Tests must also assess long-term effects because short-term trials do not take into account the ability of plants to recover following sublethal application doses of an herbicide, nor do they adequately assess reproduction, which has been shown to be a more sensitive endpoint than biomass in many cases. For annual species which fully rely on yearly seed production to ensure their persistence within a community, assessment of herbicide sensitivity at sublethal doses would be more accurately estimated through the effects on reproduction rather than solely on biomass.

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Conflict of interest The authors declare that they have no conflict of interest.

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Annex 1

Non-linear regression logistic models used to calculate:

IC50 (xmid)

$$\frac{Asym}{1+\left(\frac{logdose}{xmid}\right)^{scal}}$$

IC25 (xmid)

$$\frac{Asym}{1+\left(\frac{0.25}{0.75}\right)\left(\frac{logdose}{xmid}\right)^{scal}}$$

IC10 (xmid)

$$\frac{Asym}{1+\left(\frac{0.1}{0.9}\right)\left(\frac{logdose}{xmid}\right)^{scal}}$$

Where *Asym* is the asymptote of the model and is forced to be the mean biomass (or total seed biomass or number of seeds) of the controls; *xmid* represents the inhibition concentration dose (IC) and *scal* is the parameter referred to the slope of the graph defined by the model *logdose* is the variable dose of herbicide entered as logarithm (dose + 1).

Annex 2

Table A2.1: Summary of the IC50s (defined as the dosage that resulted in a 50 % reduction in the overall aboveground biomass) for species tested with tribenuron and 2,4-D. The label rate for tribenuron was 7.5 g acid-equivalent (a.e.) ha^{-1} and for 2,4-D was 564 g a.e. ha^{-1}. ‘ST’ indicates the short-term and ‘LT’ the long-term harvest data. We used ‘>7.500’ and ‘>564.000’ when the estimated IC50s were greater than the highest dose evaluated for each herbicide. ‘NE’ indicates no statistically significant effect. The rarity of each species is indicated as [R] rare or [C] common.

	Species	Test duration	IC50 (g a.e. ha^{-1})	95 % Confidence Intervals	% of label rate
Tribenuron					
Apiaceae	*Bupleurum rotundifolium* [R]	ST	3.911	2.744 - 5.441	52.144
		LT	3.215	2.126 - 4.685	42.872
	Scandix pecten-veneris [C]	ST	>7.500		
		LT	>7.500		
Brassicaceae	*Neslia paniculata* [R]	ST	NE		
		LT	NE		
	Rapistrum rugosum [C]	ST	0.982	0.743 - 1.255	13.099
		LT	NE		
Papaveraceae	*Papaver argemone* [R]	ST	0.292	0.255 - 0.331	3.898
		LT	0.436	0.368 - 0.507	5.812
	Papaver rhoeas [C]	ST	1.100	0.691 - 1.608	14.665
		LT	1.674	1.091 - 2.419	22.320
Rubiaceae	*Asperula arvensis* [R]	ST	NE		
		LT	NE		
	Galium spurium [C]	ST	>7.500		
		LT	>7.500		
2,4-D					
Apiaceae	*Bupleurum rotundifolium* [R]	ST	>564.000		
		LT	>564.000		
	Scandix pecten-veneris [C]	ST	389.994	290.089 - 524.188	69.148
		LT	489.908	212.010 - 1130.358	86.863
Brassicaceae	*Neslia paniculata* [R]	ST	>564.000		
		LT	433.510	354.435 - 530.177	76.864
	Rapistrum rugosum [C]	ST	184.349	156.050 - 217.747	32.686
		LT	>564.000		
Papaveraceae	*Papaver argemone* [R]	ST	>564.000		
		LT	>564.000		
	Papaver rhoeas [C]	ST	>564.000		
		LT	>564.000		
Rubiaceae	*Asperula arvensis* [R]	ST	NE		
		LT	NE		
	Galium spurium [C]	ST	>564.000		
		LT	NE		

Table A2.2: Summary of the IC50s (defined as the dosage that resulted in a 50 % reduction in total seed biomass and number of seeds) for species tested with tribenuron and 2,4-D. The selected label rate for tribenuron was 7.5 g acid equivalent (a.e.) ha^{-1} and for 2,4-D was 564 g a.e. ha^{-1}. 'ST' indicates the short-term and 'LT' the long-term harvest data. We used '>7.500' and '>564.000' when the estimated IC50s were greater than the highest dose evaluated for each herbicide. 'NE' indicates no statistically significant effect. The rarity of each species is indicated as [R] rare or [C] common.

	Species	Test duration	Variable	IC50 (g a.e. ha^{-1})	95 % Confidence Intervals	% of label rate
Tribenuron						
Apiaceae	*Bupleurum rotundifolium* [R]	LT	Seed biomass	>7.500		
			Num of seeds	NE		
	Scandix pecten-veneris [C]	LT	Seed biomass	3.454	2.269 - 5.068	46.053
			Num of seeds	NE		
Brassicaceae	*Neslia paniculata* [R]	ST	Seed biomass	NE		
			Num of seeds	NE		
		LT	Seed biomass	NE		
			Num of seeds	NE		
Papaveraceae	*Papaver argemone* [R]	LT	Seed biomass	0.295	0.262 - 0.329	3.936
	Papaver rhoeas [C]	LT	Seed biomass	0.325	0.118 - 0.569	4.329
Rubiaceae	*Asperula arvensis* [R]	ST	Seed biomass	NE		
			Num of seeds	NE		
		LT	Seed biomass	>7.500		
			Num of seeds	NE		
	Galium spurium [C]	LT	Seed biomass	>7.500		
			Num of seeds	NE		
2,4-D						
Apiaceae	*Bupleurum rotundifolium* [R]	LT	Seed biomass	>564.000		
			Num of seeds	NE		
	Scandix pecten-veneris [C]	LT	Seed biomass	219.699	17.251 - 2667.751	38.954
			Num of seeds	NE		
Brassicaceae	*Neslia paniculata* [R]	ST	Seed biomass	166.888	126.301 - 220.415	29.590
			Num of seeds	111,746	-0.317 - 0.464	19.813
		LT	Seed biomass	353.871	239.404 - 522.840	62.743
			Num of seeds	482.727	225.121 - 1033.809	85.590
Papaveraceae	*Papaver argemone* [R]	LT	Seed biomass	435.717	60.625 - 3093.881	77.255
			Num of seeds	92.821	28.656 - 295.819	16.458
	Papaver rhoeas [C]	LT	Seed biomass	NE		
			Num of seeds	NE		
Rubiaceae	*Asperula arvensis* [R]	ST	Seed biomass	>564.000		
			Num of seeds	NE		
		LT	Seed biomass	>564.000		
			Num of seeds	NE		
	Galium spurium [C]	LT	Seed biomass	>564.000		
			Num of seeds	NE		

Annex 3

Table A3.1: Summary of the IC10s (defined as the dosage that resulted in a 10 % reduction in the overall aboveground biomass) for species tested with tribenuron and 2,4-D. The label rate for tribenuron was 7.5 g acid-equivalent (a.e.) ha^{-1} and for 2,4-D was 564 g a.e. ha^{-1}. 'ST' indicates the short-term and 'LT' the long-term harvest data. We used '>7.500' and '>564.000' when the estimated IC10s were greater than the highest dose evaluated for each herbicide. 'NE' indicates no statistically significant effect. The rarity of each species is indicated as [R] rare or [C] common.

	Species	Test duration	IC10 (g a.e. ha^{-1})	95 % Confidence Intervals	% of label rate
Tribenuron					
Apiaceae	*Bupleurum rotundifolium* [R]	ST	0.344	0.186 - 0.523	4.587
		LT	0.471	0.224 - 0.768	6.284
	Scandix pecten-veneris [C]	ST	0.507	0.223 - 0.857	6.759
		LT	1.869	1.036 - 3.044	24.926
Brassicaceae	*Neslia paniculata* [R]	ST	NE		
		LT	NE		
	Rapistrum rugosum [C]	ST	0.162	0.077 - 0.253	2.156
		LT	NE		
Papaveraceae	*Papaver argemone* [R]	ST	0.073	0.046 - 0.101	0.977
		LT	0.168	0.028 - 0.327	2.240
	Papaver rhoeas [C]	ST	0.250	0.109 - 0.410	3.339
		LT			
Rubiaceae	*Asperula arvensis* [R]	ST	NE		
		LT	NE		
	Galium spurium [C]	ST	0.590	0.177 - 1.148	7.873
		LT	2.815	0.901 - 6.655	37.534
2,4-D					
Apiaceae	*Bupleurum rotundifolium* [R]	ST	141.594	81.704 - 244.851	25.105
		LT	144.747	84,338 - 247.918	25.664
	Scandix pecten-veneris [C]	ST	84.349	51.541 - 137.644	14.956
		LT	52.567	29.266 - 93.807	9.320
Brassicaceae	*Neslia paniculata* [R]	ST	72.978	49.799 - 106.733	12.939
		LT	94.969	67.199 - 134.046	16.838
	Rapistrum rugosum [C]	ST	36.350	26.950 - 48.911	6.445
		LT	39.926	18.870 - 83.294	7.079
Papaveraceae	*Papaver argemone* [R]	ST	101.351	65.157 - 157.344	17.970
		LT	19.583	5.450 - 64.677	3.472
	Papaver rhoeas [C]	ST	34.237	1.097 - 591.096	6.070
		LT	24.421	-0.936 - 10118.056	4.330
Rubiaceae	*Asperula arvensis* [R]	ST	NE		
		LT	NE		
	Galium spurium [C]	ST	6.365	0.445 - 36.546	1.129
		LT	NE		

Table A3.2: Summary of the IC10s (defined as the dosage that resulted in a 10 % reduction in total seed biomass and number of seeds) for species tested with tribenuron and 2,4-D. The selected label rate for tribenuron was 7.5 g acid-equivalent (a.e.) ha^{-1} and for 2,4-D was 564 g a.e. ha^{-1}. 'ST' indicates the short-term and 'LT' the long-term harvest data. We used '>7.500' and '>564.000' when the estimated IC10s were greater than the highest dose evaluated for each herbicide. 'NE' indicates no statistically significant effect. The rarity of each species is indicated as [R] rare or [C] common.

	Species	Test duration	Variable	IC10 (g a.e. ha^{-1})	95 % Confidence Intervals	% of label rate
Tribenuron						
Apiaceae	*Bupleurum rotundifolium* [R]	LT	Seed biomass	0.003	-0.041 - 0.050	0.042
			Num of seeds	NE		
	Scandix pecten-veneris [C]	LT	Seed biomass	1.070	0.440 - 1.976	14.270
			Num of seeds	NE		
Brassicaceae	*Neslia paniculata* [R]	ST	Seed biomass	NE		
			Num of seeds	NE		
		LT	Seed biomass	NE		
			Num of seeds	NE		
Papaveraceae	*Papaver argemone* [R]	LT	Seed biomass	0.175	0.147 - 0.204	2.337
	Papaver rhoeas [C]	LT	Seed biomass	0.051	-0.078 - 0.199	0.686
Rubiaceae	*Asperula arvensis* [R]	ST	Seed biomass	NE		
			Num of seeds	NE		
		LT	Seed biomass	>7.500		
			Num of seeds	NE		
	Galium spurium [C]	LT	Seed biomass	2.081	1.046 - 3.638	27.743
			Num of seeds	NE		
2,4-D						
Apiaceae	*Bupleurum rotundifolium* [R]	LT	Seed biomass	8.328	-0.946 - 1609.438	1.477
			Num of seeds	NE		
	Scandix pecten-veneris [C]	LT	Seed biomass	35.855	9.202 - 132.142	6.357
			Num of seeds	NE		
Brassicaceae	*Neslia paniculata* [R]	ST	Seed biomass	61.633	36.833 - 102.689	10.928
			Num of seeds	4.152	0.207 - 20.986	0.736
		LT	Seed biomass	101.259	52.438 - 194.681	17.954
			Num of seeds	70.335	25.610 - 190.232	12.471
Papaveraceae	*Papaver argemone* [R]	LT	Seed biomass	4.954	0.093 - 31.432	0.878
			Num of seeds	3.384	-0.217 - 23.549	0.600
	Papaver rhoeas [C]	LT	Seed biomass	NE		
			Num of seeds	NE		
Rubiaceae	*Asperula arvensis* [R]	ST	Seed biomass	284.627	59.582 - 1345.654	50.466
			Num of seeds	NE		
		LT	Seed biomass	143.844	56.115 - 366.326	25.504
			Num of seeds	NE		
	Galium spurium [C]	LT	Seed biomass	>564.000		
			Num of seeds	NE		

Capítol 4

Modulation of the competitive relationship between wheat and the characteristic and rare arable species of winter cereal fields by fertilizers

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L'editor i els revisors han recomanat la publicació de l'article a *Weed Research*
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RESUM

Modulació de la competència entre el blat i les espècies arvenses característiques i rares dels cultius cerealistes de secà pels fertilitzants

Algunes espècies arvenses característiques dels hàbitats cerealistes de secà (o espècies segetals) són molt poc freqüents i requereixen una atenció especial pel que fa a la seva conservació. En conseqüència, resulta fonamental conèixer quin és l’efecte de les pràctiques agrícoles sobre les seves poblacions. El cultiu i les espècies arvenses competeixen pels mateixos recursos, principalment nutrients, aigua i llum, i per aquest motiu tendeixen a ser mútuament excloents. Així doncs, les pràctiques agrícoles que promouen la producció del cultiu mitjançant l’increment de la seva capacitat competitiva minimitzen el desenvolupament de les poblacions d’espècies arvenses. L’aplicació de fertilitzants es considera fonamental a l’hora de determinar les relacions de competència entre el cultiu i les espècies arvenses, ja que sol estimular el creixement del cultiu i, per tant, incrementa la competència per la llum. Per aquest motiu es va avaluar la resposta de les espècies segetals segons el tipus (orgànic i mineral) i la dosi (alta i baixa) de fertilització en competència amb el cereal o no mitjançant un experiment en condicions controlades. Aquest experiment es va dur a terme amb quatre parelles d’espècies segetals, una més rara i una més comuna, que pertanyen a famílies diferents. En particular, 1) s’ha avaluat l’efecte competitiu del blat sobre el creixement de les plantes segetals comunes i rares i com aquesta relació de competència és modulada pel tipus i la dosi de fertilització, 2) s’ha determinat l’efecte de diferents dosis i tipus de fertilització sobre el moment de la floració de les espècies arvenses i si aquesta relació està condicionada per la competència amb el cereal i 3) s’ha avaluat si la capacitat competitiva de les espècies rares difereix de les més comunes emparentades filogenèticament.

El fertilitzant mineral i el fertilitzant orgànic compostat es van aplicar a dues dosis diferents, però equivalents entre tipus de fertilitzants, en testos amb un volum constant de substrat. En un grup d’aquests testos s’hi va disposar una sola planta arvense al centre, mentre que en un altre grup de testos s’hi van sembrar, a més, dues línies de cinc plantes de blat cadascuna. Es va avaluar l’efecte de la fertilització i de la competència amb el blat sobre les espècies arvenses (biomassa aèria total i reproductiva i longitud de les tiges o llargada de fulles) així com els efectes sobre l’alçada del blat utilitzant models lineals mixtes. També es va determinar l’efecte de la fertilització i la competència en el moment de floració de les espècies arvenses mitjançant models de riscos proporcionals mixtes (regressió de Cox d’efectes mixtes).

Les espècies arvenses característiques van créixer més amb fertilització mineral alta quan creixien soles. Però en competència amb el blat ni la fertilització mineral ni les dosis altes de fertilitzants van afavorir el seu creixement. El blat, en canvi, va créixer més amb fertilització mineral alta amb independència de la presència de les espècies arvenses. El cereal és més eficient que les espècies segetals a l'hora de captar els nutrients del sòl (Robinson & Sutherland 2002), de manera que davant d'una disponibilitat de nutrients més alta creix més ràpidament, el que li confereix un avantatge competitiu per la llum (Goldberg & Miller 1990; Weiner et al. 2001; Tang et al. 2014). La fertilització orgànica, per contra, permet a les espècies segetals créixer més, ja que redueix la competència exercida pel cultiu. El tipus i la dosi de fertilització van afectar la mida de les plantes segetals de la mateixa manera tant si creixien en solitari com en competència amb el blat. A dosis elevades de fertilitzants minerals el cereal monopolitza l'accés a la llum. Això provoca que les espècies segetals, malgrat acumulen menys biomassa en competència amb el blat, són capaces d'assolir mides semblants a quan creixen soles per competir amb el cereal per la llum (Kleijn & van der Voort 1997).

La floració de les espècies testades va tendir a ser més tardana a dosis baixes de fertilització (sis de vuit) i amb fertilització orgànica (totes les espècies), tot i que els efectes van ser estadísticament significatius només per algunes d'elles. Una floració més tardana pot comportar una producció de llavors més baixa, fet que pot afectar negativament la mida de les poblacions en el futur (Goldberg & Miller 1990; Storkey 2006; Fried et al. 2012). Tot i això, la fertilització i la competència amb el cultiu van repercutir menys en el moment de floració que en la biomassa.

Dues de les espècies segetals rares van ser més sensibles a la competència amb el blat que les seves parelles comunes en termes de creixement (biomassa i mida). Malgrat això, no totes les parelles d'espècies van respondre de la mateixa manera davant la competència amb el cereal sota diferents dosis i tipus de fertilització. En termes generals, el cereal és menys competitiu davant les espècies segetals quan la fertilització és orgànica. Per aquest motiu la fertilització orgànica pot ser una bona solució per afavorir el desenvolupament de les espècies segetals sense perjudicar seriosament el creixement del cultiu. Els resultats d'aquest estudi suggereixen que l'efecte competitiu del cereal sota fertilització mineral a dosis elevades pot haver tingut un paper important en la davallada de la diversitat vegetal als sistemes agrícoles. Malgrat tot, la baixa freqüència d'algunes espècies segetals no es pot atribuir solament a aquest efecte, ja que no totes les espècies rares testades van respondre de la mateixa manera en comparació amb la comuna. Això indica que altres factors han contribuït en la reducció de les poblacions d'espècies segetals o en determinar-ne la seva raresa localment.

Modulation of the competitive relationship between wheat and the characteristic and rare arable species of winter cereal fields by fertilizers

Summary

Fertilizer application is considered crucial for determining the crop-weed competition relationship by stimulating plant growth and increasing the competition for belowground (soil nutrients) and aboveground resources (e.g., light and space). Characteristic arable species have suffered from the intensification of agricultural practices so much that some have become rare. In this study, we aim to assess the competitive ability of common and rare characteristic arable species when growing with wheat and how this competitive relationship is modulated by the type (organic and mineral) and dose (low or high) of fertilization in a pot experiment.

Characteristic arable species grew better at higher doses of fertilizers when growing alone. However, when growing with wheat, they were outcompeted by the cereal plants. Organic fertilization allowed characteristic arable weeds to fare better by decreasing crop competitiveness. Overall, flowering was not affected by the competition of wheat under the different fertilizer regimes. Only the onset of flowering of some characteristic weeds tended to be delayed by crop competition. Some rare species were more affected by the competition of wheat, indicating that they may be disadvantaged in a community context. However, this is not a general result, which highlights that competitiveness alone is a poor predictor of the rarity of characteristic arable species.

Keywords

Fertilizer dose; flowering date; mineral fertilizer; organic fertilizer, plant biomass; plant size.

Introduction

Crop and weed species compete for the same resources, mainly nutrients, water and light. Hence, they tend to be mutually exclusive (Tuor & Froud-Williams, 2002). Individuals that emerge earlier or are able to uptake nutrients faster may take advantage of the resources and overgrow their neighbours (Goldberg & Miller, 1990), eventually prevailing in the community at the expense of outcompeted individuals (Pyšek & Lepš, 1991; Tang et al., 2014).

Nutrient availability depends on the amount and type of fertilizer applications. Therefore, it is considered an important factor shaping the competitive interactions between crops and weeds. For instance, contrasting fertilizers such as mineral fertilizers, which rapidly release available nutrients for plant absorption, and composted organic fertilizers, which release the nutrients slowly, may have very different effects on this competitive relationship (Romanyà et al., 2012).

Farming practices enhance crop production by means of increasing crop competitive ability while minimizing weed populations. However, only a small percentage of weeds cause significant crop losses. There are some arable species that do not cause any crop production losses, yet have suffered a strong decline in abundance caused by intensive farming practices (Fried et al., 2009; Storkey et al., 2012). Most of these species thrive almost exclusively in arable fields because they depend on regular soil disturbance (i.e., tillage) and cannot succeed in more competitive habitats. Most arable weeds have an annual cycle that mimics the life cycle of the crop they are associated with. These species are considered characteristic species of arable fields (Rotchés-Ribalta et al., 2014).

Characteristic arable weeds, which particularly thrive in winter cereal fields, have adapted to cope with the competitive pressure exerted by crops. Thus, they have evolved to avoid being suppressed by the crop (Goldberg & Landa, 1991). However, these species have been greatly affected by the intensification of agricultural management, which has caused them to become rare or even locally extinct (Baessler & Klotz, 2006). That is why organic farming practices, which are related with low intensity management, encourage the presence of characteristic arable species (José-María et al., 2010). Therefore, lower fertilization rates and the application of organic fertilizers instead of mineral fertilizers among other practices can be key factors affecting the conservation of characteristic arable species, particularly of those that are rare.

Here we aim to assess the competitive response of characteristic arable species when growing with wheat, as well as how this interaction is modulated by the type (organic and mineral) and the dose (low or high) of fertilization. We performed an additive design (Goldberg & Fleetwood, 1987; Park, Benjamin & Watkinson, 2003) in a pot experiment by growing an individual of a target weed species either jointly with wheat plants or alone. We tested the competitive response of eight characteristic arable weed species from four families, using one rare and one more common species of each family. Specifically, we 1) ascertained the competitive effect of wheat plants on the growth of characteristic weed species and how this relationship is modulated by the type and the dose of fertilization, 2) determined whether the flowering onset of weed species is influenced by the competition with wheat based on the different fertilization rates and types and 3) assessed whether the competitive ability of rare arable weeds differs from the more common species.

Despite the difficulty of extrapolating results directly from pot experiments to understand the functioning of complex arable weed communities, pot-based competition experiments allow better control of biotic and abiotic factors, which help to more precisely determine species interactions (Gibson et al., 1999). We consider these types of experiments essential for achieving a comprehensive understanding of the mechanisms of competition and how they may be altered with specific agronomic practices. This then provides a sound knowledge of these mechanisms, which can be used to implement appropriate conservation measures for rare arable weed populations.

Materials and Methods

Experimental design

The experiment was conducted in 2012 at the experimental fields of the University of Barcelona, Spain. Average (± standard error) temperature during the experiment was 15.7ºC (± 2.2), ranging from 0.5 to 33.2ºC and mean relative humidity was 63.1 % (±

2.4), ranging from 10.0 to 98.8 %. Low-density polyethylene bags of 26 cm diameter and 35 cm height were used as pots containing 18 L of a non-fertilized mixture of 50 % peat and 50 % sand. Bags were drilled at the bottom to allow water drainage.

The substrate was fertilized with mineral or organic fertilizers at two different doses (low and high). For the mineral fertilization, NPK 15-15-15 product was added at 2.9 g per pot for the high dose and 1.45 g per pot for the low dose. Sieved composted farmyard manure, used as the organic fertilization, was added to the corresponding pots at a rate of 100 g for the high fertilization dose and 50 g per pot for the low dose. Both types of fertilizers represented an addition of 87 kg N ha^{-1} for the high dose and 43.5 kg N ha^{-1} for the low dose. Given the differences in the nutrient release between the mineral and organic fertilizers, the doses were adjusted to have similar nitrogen availabilities over the season.

The target weed species were all annual plants which almost exclusively inhabit arable cereal fields and are considered characteristic arable species (Rotchés-Ribalta et al., 2014). We selected eight characteristic arable species belonging to four families, from which one was rare and the other was a more common species (Table 1). The rarity of the target species was established according to their frequency in an extensive survey from north-eastern Spain (Rotchés-Ribalta et al., 2014). Weed seeds were collected in the spring-summer of 2011 from selected natural plant populations.

Table 1: Pairs of common and rare target arable weed species. The rarity status is in accordance to the frequency of occurrence of these species in a large field survey (1957 samples at the edges of 304 organic fields) from north-eastern Spain (Rotchés-Ribalta et al., 2014).

Family	Species	Frequency (%)	Rarity
Apiaceae	*Bifora testiculata* M. Spreng	0.66	Rare
	Scandix pecten-veneris L. subsp. *pecten-veneris*	15.79	Common
Brassicaceae	*Neslia paniculata* (L.) Desv. subsp. *thracica* (Velen.) Bornm.	1.33	Rare
	Rapistrum rugosum (L.) All.	8.02	Common
Papaveraceae	*Papaver argemone* L.	0.72	Rare
	Papaver rhoeas L.	61.01	Common
Rubiaceae	*Asperula arvensis* L.	0.77	Rare
	Galium aparine subsp. *spurium* (L.) Simonk	15.07	Common

Seeds were kept wet on top of trays filled with non-fertilized substrate in complete darkness at 2-4ºC for a week. Then, the trays were moved into a growth chamber with ten hours daylight at 20ºC and 14 hours night at 10ºC to promote germination. Seedlings with fully expanded cotyledons that were similar in size were individually transplanted in the centre of each pot (18th and 20th of January). Dead seedlings (62 in total) were replaced during the first 20 days. For each species, one full set of plants under all of the fertilization conditions was grown alone. Another set was sown together with ten winter wheat plants (*Triticum aestivum* var. Montcada), representing 200 seeds m^{-2} to address the effect of crop-weed competition. Wheat seeds were sown the 17th and 18th of January at 1.5 cm depth along two lines in each pot, which were separated by 12.5 cm. This simulated the sowing lines experienced under field conditions. Pots were watered regularly.

All of the pots were placed on a grid and distributed in five blocks, corresponding to replicates. Within each block, two sub-blocks were determined according to the different levels of fertilization. We also included twenty extra pots sown only with wheat under the different fertilization conditions.

Data collection

Different measures have been used to evaluate the competitive performance of plants, with biomass measures being the most commonly used method for assessing the competitiveness between plant species. Plant material was harvested when the wheat was dry and had hard kernels (4th of June), almost five months after sowing. The aboveground biomass of both crop and weed plants was cut at the soil level, bagged separately, dried at 60ºC for at least five days and weighed afterwards.

However, when light is the main source of plant competition, plant size may also be relevant (Goldberg & Fleetwood, 1987). The sum of the length of the main stems of the species with ascending leafy stems (*A. arvensis* and *G. aparine* subsp. *spurium*, hereinafter *G. spurium*) was measured. The mean lengths of the three larger rosette leaves were measured for the other species. The heights of four randomly selected wheat tillers in each pot were measured to obtain the mean height of the wheat plants. These measurements were carried out at four different moments throughout the duration of the experiment.

The persistence of characteristic arable species relies heavily on reproduction to ensure future populations. Therefore, the analysis of the effects of competition on flowering is crucial. The phenology of the weeds, i.e., vegetative or flowering, was also recorded regularly, up to seven times.

Statistical analysis

- *Effects of fertilization and wheat competition on weed growth*. Mixed effect models were used to assess the effects of the type of fertilization, dose of fertilization, wheat competition and their interactions on the aboveground biomass. The plant biomass was log transformed to achieve normality and homoscedasticity of the residuals. When the homoscedasticity conditions were not met, we included the heteroscedasticity structure in the error term. Blocks and sub-blocks (nested to block) were included as random factors in the model.

To evaluate the differences between the rare and the common species of a given family, we fitted the mixed effect models to the aboveground biomass jointly for each pair of species within each family. The factor rarity was then included in the model to assess whether the difference between the two species was significant.

Similar models were also run for each weed species to assess the effects of wheat competition on the size of the plants under different fertilizations (total stem length or mean rosette leaf length, depending on the species) and over time. As these measures were evaluated four times during the experiment, the sampling date was included in the models through a polynomial trend (linear, quadratic and cubic terms). This allowed for modelling of the temporal trend as best as possible without relying on complicated growth models, and enabled the detection of differences on the final sizes and the growth rates between species. The total stem length and the mean rosette leaf length were log transformed to achieve normality and homoscedasticity of the residuals. When the homoscedasticity requirement was not met, the models were fitted, including a heteroscedastic error structure.

For all of the analyses we used the R 3.0.3 program (R Development Core Team, 2014) with the nlme package (Pinheiro et al., 2013).

- *Effects of fertilization and weed competition on wheat growth*. We also assessed the competitive effects of the weeds on the mean height of the wheat plants growing under different fertilizer regimes. We used a mixed effect model, including the weed species, the type and the dose of fertilization, the sampling date following the same approach as for weed growth, and their interactions as fixed factors. The mean wheat height was log-transformed to achieve normality and homoscedasticity. Blocks and sub-blocks (nested to block) were included as random factors in the model. These analyses were carried out with R 3.0.3 (R Development Core Team, 2014) using the nlme package (Pinheiro et al., 2013).

- *Effects of fertilization and wheat competition on date of flowering*. We assessed the effects of the types and the doses of fertilizers, as well as the wheat competition, on the flowering time of each weed species. We used the Cox proportional hazards mixed effects models to enable inclusion of the sub-block and block random effects. This analysis was carried out using R 3.0.3 (R Development Core Team, 2014) with the coxme package (Therneau, 2012).

Results

Effects of fertilization and wheat density on weed growth

Overall, the aboveground biomass of the characteristic arable species was lower when growing with wheat (Table 2). They grew bigger at high fertilization doses, but less vigorously when the fertilization was organic, compared to mineral fertilization. However, for all of the species other than *R. rugosum,* the effect of organic fertilization was significant only when growing without wheat. Similarly, for some species (*A. arvensis*, *G. spurium*, *B. testiculata*, *P. argemone* and *P. rhoeas*), high doses of fertilization led to higher biomasses only when growing alone. High doses of mineral fertilizer induced a larger biomass of *G. spurium* and *B. testiculata,* but this trend was not observed under organic fertilization.

Table 2: Effects of the fixed factors on the biomass of characteristic arable plants. Estimated coefficients and their standard errors for the mixed effect models are presented. Significance of the effects is indicated as · when p-value < 0.1, * <0.05; ** < 0.01 and *** < 0.001. Significant differences (p-value < 0.05) in each parameter between the rare (R) and the common (C) species of each family are shaded.

	Bifora testiculata (R)	*Scandix pecten-veneris* (C)	*Neslia paniculata* (R)	*Rapistrum rugosum* (C)
Intercept	0.639 ± 0.101***	1.945 ± 0.308***	1.276 ± 0.134***	4.343 ± 0.569***
Type of fertilizer				
Organic (vs. Mineral)	-0.290 ± 0.132*	-1.097 ± 0.405*	-0.648 ± 0.155**	-2.480 ± 0.711**
Dose of fertilizer				
High fertilizer (vs. Low fertilizer)	0.721 ± 0.132***	1.121 ± 0.405*	0.849 ± 0.155***	0.278 ± 0.711
Sowing wheat density				
Wheat (vs. No wheat)	-0.569 ± 0.132***	-1.443 ± 0.405**	-0.975 ± 0.155***	-2.165 ± 0.711**
Organic fertilizer (vs. Mineral) × High fertilizer (vs. Low)	-0.651 ± 0.154**	-0.334 ± 0.472	-0.352 ± 0.174•	0.572 ± 0.821
Organic fertilizer (vs. Mineral) × Wheat (vs. No wheat)	0.526 ± 0.154**	1.146 ± 0.472*	0.706 ± 0.174***	0.766 ± 0.821
High fertilizer (vs. Low) × Wheat (vs. No wheat)	-0.337 ± 0.154*	-0.751 ± 0.472	-0.365 ± 0.174•	-0.512 ± 0.821

	Papaver argemone (R)	*Papaver rhoeas* (C)	*Asperula arvensis* (R)	*Galium spurium* (C)
Intercept	0.661 ± 0.156***	1.673 ± 0.331***	1.321 ± 0.226***	1.348 ± 0.144***
Type of fertilizer				
Organic (vs. Mineral)	-0.236 ± 0.183	-1.407 ± 0.335**	-0.998 ± 0.236**	-0.983 ± 0.175***
Dose of fertilizer				
High fertilizer (vs. Low fertilizer)	0.955 ± 0.183***	0.699 ± 0.253*	0.746 ± 0.239**	0.713 ± 0.175**
Sowing wheat density				
Wheat (vs. No wheat)	-0.733 ± 0.191**	-1.637 ± 0.331***	-1.123 ± 0.231***	-1.277 ± 0.175***
Organic fertilizer (vs. Mineral) × High fertilizer (vs. Low)	-0.446 ± 0.216•	0.020 ± 0.045	-0.088 ± 0.156	-0.454 ± 0.202*
Organic fertilizer (vs. Mineral) × Wheat (vs. No wheat)	0.462 ± 0.217*	1.412 ± 0.335***	0.931 ± 0.242**	1.137 ± 0.202***
High fertilizer (vs. Low) × Wheat (vs. No wheat)	-0.656 ± 0.217**	-0.688 ± 0.252*	-0.534 ± 0.221*	-0.492 ± 0.202*

The rarity status did not show a general correlation with the growth pattern of weeds. For instance, the rare species of Apiaceae and Brassicaceae grew less vigorously than their respective common species, while no differences were found for Papaveraceae and Rubiaceae (Table 2).

The size of the characteristic arable plants followed the same pattern as the biomass results. Weeds growing with wheat were shorter than when growing alone, as well as under organic fertilization and at lower fertilization doses (Fig. 1). However, the interactions between wheat competition and the type and the dose of fertilizers were generally not significant (Appendix S1). This indicates that fertilizers affected the plant growth independently of the presence of wheat, which differs from the biomass results. Regarding the rarity status, the rare species *B. testiculata*, *P. argemone* and *A. arvensis* grew shorter than their respective common species. Nevertheless, their response to the tested variables was similar to the common species because no significant interactions were found (Appendix S1).

Wheat growth

Wheat plants grew taller with higher doses of fertilizer inputs, particularly when mineral fertilizer was applied. This is indicated by the significant effect of the interaction between the type and the dose of fertilization (Table 3). Characteristic arable species growing with wheat did not significantly affect the height of wheat at any of the doses and fertilizer types.

Effects of fertilization and wheat competition on flowering onset

Organic fertilization led to later flowering in general, but the differences were only marginally significant for the rare species *A. arvensis* and *B. testiculata*. Lower fertilization rates also led to later flowering for six out of eight species, but the effect was statistically significant only for *P. rhoeas* (Table 4). Rare species showed no significant differences with respect to their common counterparts in terms of the flowering onset, except for *B. testiculata*, which flowered significantly later than its paired common species (Table 4).

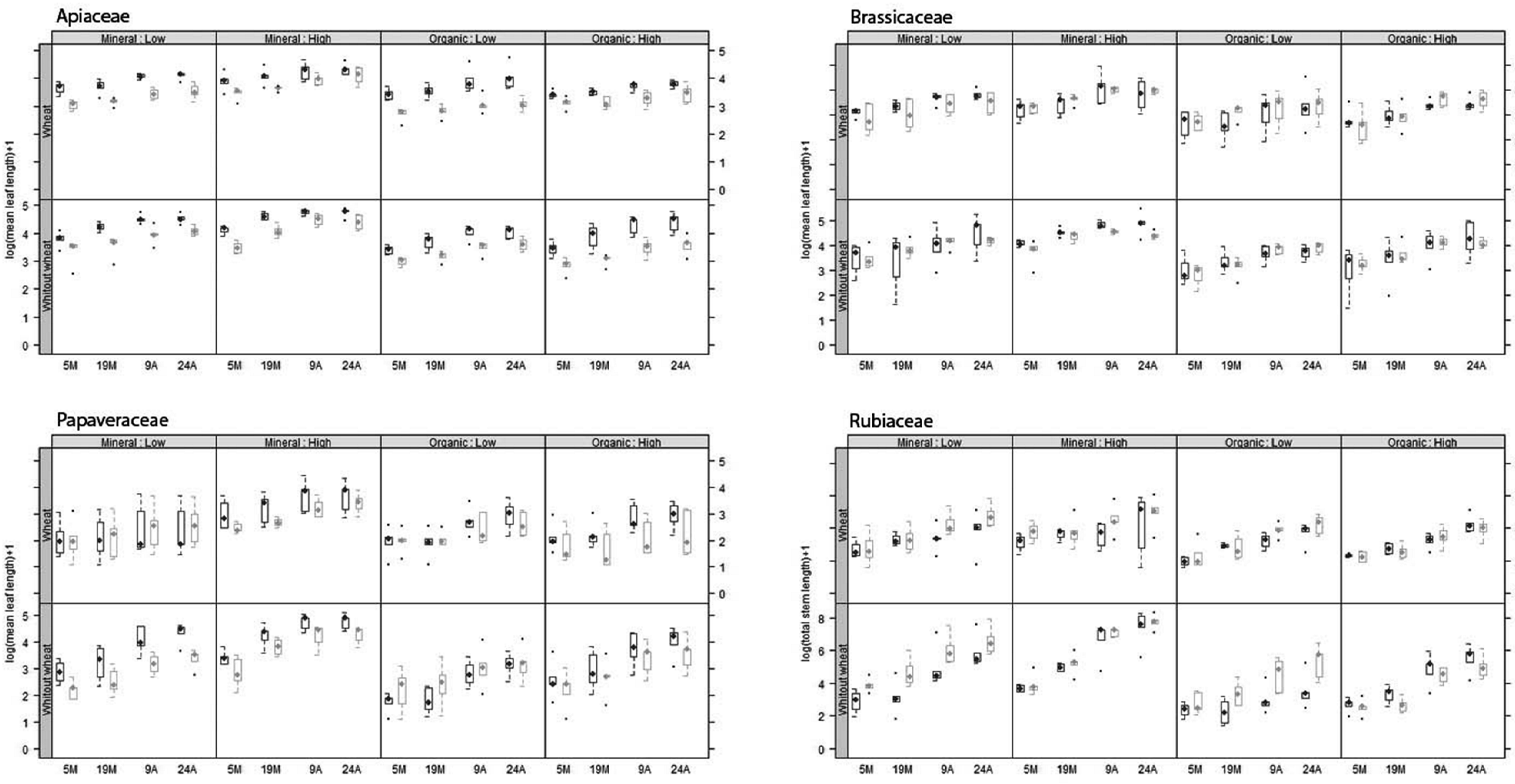

Fig. 1: Box-plots for the average rosette leaf length (Apiaceae, Brassicaceae and Papaveraceae) and the total stem height (Rubiaceae) for the common (dark grey) and rare (light grey) species of each family at each sampling date (5th March, 19th March, 9th April and 24th April). 'Wheat' and 'Without wheat' indicate the condition of growing together with or without wheat respectively; doses of fertilizer are shown as 'High' and 'Low'; and 'Mineral' and 'Organic' represent the type of fertilizer applied.

Table 3: Effects of the fixed factors on the mean height of wheat plants. Estimated coefficients and their standard errors for the mixed effect models are presented. Significance of the effects is indicated as · when p-value < 0.1, * <0.05; ** < 0.01 and *** < 0.001. Rarity status of arable weed species is indicated as (R) and (C) for rare and common species respectively.

	Estimate ± SE
Type of fertilizer	
Organic (vs. Mineral)	-35.551 ± 5.011***
Dose of fertilizer	
High fertilizer (vs. Low fertilizer)	32.986 ± 5.129***
Sampling date	
Sampling date (Linear term)	320.368 ± 4.653***
Sampling date (Quadratic term)	174.272 ± 3.865***
Sampling date (Cubic term)	47.788 ± 2.873***
Organic fertilizer (vs. Mineral) × High fertilizer (vs. Low)	-24.412 ± 6.739**
Organic fertilizer (vs. Mineral) × Sampling date L	-53.483 ± 5.195***
Organic fertilizer (vs. Mineral) × Sampling date Q	-22.628 ± 4.334***
Organic fertilizer (vs. Mineral) × Sampling date C	-10.029 ± 3.257**
High fertilizer (vs. Low) × Sampling date L	0.925 ± 5.099
High fertilizer (vs. Low) × Sampling date Q	-1.011 ± 4.256
High fertilizer (vs. Low) × Sampling date C	1.374 ± 3.196
Growing with:	
Bifora testiculata (R) (vs. Alone)	-4.984 ± 2.728•
*Scandix pecten-veneri*s (C) (vs. Alone)	-1.047 ± 2.773
Neslia paniculata (R) (vs. Alone)	-0.856 ± 2.775
Rapistrum rugosum (C) (vs. Alone)	2.014 ± 2.809
Papaver argemone (R) (vs. Alone)	0.060 ± 2.786
Papaver rhoeas (C) (vs. Alone)	0.741 ± 2.794
Asperula arvensis (R) (vs. Alone)	-3.529 ± 2.745
Galium spurium (C) (vs. Alone)	-4.831 ± 2.730•

Table 4: Effects of the fixed factors on the time to flowering of characteristic arable plants. Estimated coefficients and their standard errors from the Cox mixed effect models are presented. Significance of the effects is indicated as · when p-value < 0.1, * <0.05; ** < 0.01 and *** < 0.001. Significant differences (p-value < 0.05) in each parameter between the rare (R) and the common (C) species of each family are shaded.

	Apiaceae		Brassicaceae	
	Bifora testiculata (R)	*Scandix pecten-veneris* (C)	*Neslia paniculata* (R)	*Rapistrum rugosum* (C)
Intercept	-	- [a]	-	-
Type of fertilizer				
Organic (vs. Mineral)	-1.049 ± 0.591·	-0.659 ± 0.566	-0.914 ± 0.560	0.220 ± 0.589
Dose of fertilizer				
High fertilizer (vs. Low fertilizer)	0.168 ± 0.560	-0.014 ± 0.566	0.830 ± 0.563	1.083 ± 0.614· ·
Wheat density				
Wheat (vs. No wheat)	-0.136 ± 0.559	-0.918 ± 0.574	-0.396 ± 0.553	0.156 ± 0,596
Organic fertilizer (vs. Mineral) × High fertilizer (vs. Low)	-0.128 ± 0.645	-0.332 ± 0.657	-0.635 ± 0.642	-0.349 ± 0.665
Organic fertilizer (vs. Mineral) × Wheat (vs. No wheat)	0.504 ± 0.651	0.249 ± 0.668	0.905 ± 0.645	-0.528 ± 0.680
High fertilizer (vs. Low) × Wheat (vs. No wheat)	0.504 ± 0.651	0.376 ± 0.660	-0.472 ± 0.639	-0.285 ± 0.670

	Papaveraceae		Rubiaceae	
	Papaver argemone (R)	*Papaver rhoeas* (C)	*Asperula arvensis* (R)	*Galium spurium* (C)
Intercept	-	-	-	-
Type of fertilizer				
Organic (vs. Mineral)	0.066 ± 0.564	-0.465 ± 0.593	-0.979 ± 0.572·	-0.623 ± 0.565
Dose of fertilizer				
High fertilizer (vs. Low fertilizer)	1.030 ± 0.573·	1.214 ± 0.598*	0.138 ± 0.578	-0.623 ± 0.565
Wheat density				
Wheat (vs. No wheat)	-0.087 ± 0.590	-1.165 ± 0.649·	-0.570 ± 0.551	-0.829 ± 0.565
Organic fertilizer (vs. Mineral) × High fertilizer (vs. Low)	-0.931 ± 0.688	-0.749 ± 0.691	0.624 ± 0.692	0.278 ± 0.646
Organic fertilizer (vs. Mineral) × Wheat (vs. No wheat)	0.109 ± 0.673	1.437 ± 0.727*	0.866 ± 0.644	0.484 ± 0.646
High fertilizer (vs. Low) × Wheat (vs. No wheat)	-0.169 ± 0.673	0.191 ± 0.691	-0.273 ± 0.643	0.484 ± 0.646

[a] Cox mixed effect models do not provide a baseline hazard. Estimated magnitude of the difference was -2.859 ± 0.669***.

Discussion

Competitive relationships between wheat and weeds

Weed growth (in terms of aboveground biomass) responded positively to higher soil nutrient levels when growing alone, as noted previously (Andreasen, Litz & Streibig, 2006). In most cases, however, the positive effect of the higher fertilization rates on the growth of arable weeds was observed only under mineral fertilization. Composted manure has fewer nutrients immediately available to plants than do mineral fertilizers because of a slower release (Romanyà et al., 2012). This difference led to both smaller arable weeds and smaller wheat plants. This effect was more notorious for the wheat than for the weed species, but it may vary depending on the responses of arable species to the different nutrient availabilities (Liebman et al., 2004; Blackshaw, 2005).

However, the different rates of organic and mineral fertilization tested did not affect the weed biomass when growing in competition with the wheat plants. Similar results were previously found for more generalist weeds (Andreasen et al., 2006). Although characteristic arable species were able to grow better under higher fertilization doses, their growth was dramatically reduced by wheat competition because wheat uses nutrients more efficiently. Therefore, crops outcompete the characteristic arable species by pre-empting available soil nutrients and limiting access to light (Goldberg & Miller, 1990; Robinson & Sutherland, 2002; Tang et al., 2014). Other studies have even found weed biomass to be reduced when increasing fertilization, which indicates the importance of fertilization on stimulating crop growth and reducing weed growth (Kleijn & van der Voort, 1997).

Contrarily, the type and the dose of the fertilization had the same effect on the size (total stem length or mean rosette leaf length) of the characteristic arable weeds when growing alone or with wheat. At higher fertilization rates, especially in mineral fertilizer, wheat outcompeted characteristic arable weeds for light (Pyšek & Lepš, 1991). In response to this, characteristic weed plants grew as tall or produced rosettes as large as when growing alone to absorb light but without increasing their biomass (Kleijn & van der Voort, 1997). However, not all arable species fit this pattern. Some generalist weeds are good competitors and can take advantage of the added fertilization despite competing with a crop (Goldberg & Miller, 1990; Blackshaw, 2005; Blackshaw & Brandt, 2009).

The analysis suggests that the competitive effect of the wheat is higher than that of the characteristic arable weeds studied. Therefore, none of the arable species tested had a significant effect on the growth of wheat. This trend might seem skewed because there was only one weed against several wheat plants. However, that ratio is commonly found in the fields. Wheat plants were able to uptake nutrients faster than the characteristic arable weeds (Tang et al., 2014) and thus grew taller, as previously noted (Kleijn & van der Voort, 1997). This performance is particularly remarkable at higher rates of mineral fertilization, as it represents more immediate available nutrients for plants.

On the contrary, organic fertilization, independent of the amount applied, gave rise to remarkably smaller wheat plants than mineral fertilization. Moreover, the differences were larger for the wheat than for the characteristic arable species. Therefore, organic fertilizers may allow characteristic and also rare arable species to thrive in organically fertilized fields, because of reduced crop competitiveness.

Relative competitive ability of common and rare arable species

The fertilization can affect the relative competitive ability of the weeds growing with crops. Therefore, we expected that the species currently considered rare to have undergone more severe declines than common species over the past few decades, as a result of the high fertilization rates that have characterized agricultural management (Pyšek & Lepš, 1991; Tilman et al., 2002). In our study, the organic fertilization, where fewer nutrients were available, generally led to less vigorous weeds. However, *B. testiculata* and *N. paniculata*, the rare Apiaceae and Brassicaceae species, respectively, experienced this effect significantly more than their paired common species. This indicates that these rare species may be at a disadvantage in a community context, as demonstrated by other rare arable species (Epperlein et al., 2014). Moreover, in terms of biomass, these rare species were more outcompeted by the wheat than their paired common species. This emphasizes the relevance of the highly competitive crop species in determining the rarity of these characteristic arable weeds.

However, the effects of fertilization rates were variable among species. Among the Rubiaceae species, the rare species was better able to take advantage of higher fertilizer doses, but only when growing alone. Both Papaveraceae species responded similarly in terms of biomass. Therefore, while rarity of some characteristic arable species (such as *B. testiculata* and *N. paniculata*) might be caused by the competition with crop species under high doses of mineral fertilization, for other species, the rarity might be explained by other management practices, such as herbicide application (Rotchés-Ribalta et al., 2015).

With regard to the total stem heights and rosette leaf lengths, despite the fact that the rare species were shorter or had smaller leaves than the common species, both rare and common species exhibited the same response to wheat competition, as no significant interactions were found. This indicates that the rarity of arable weeds is not related to their size response to the different fertilizer regimes.

Reproductive competitive response of characteristic weeds

Generally the flowering onset was not significantly affected by the wheat competition at the different fertilization types and doses. A trend of later flowering was observed on some characteristic arable species at low fertilizer doses, especially under organic fertilization. However, only *P. rhoeas* flowered significantly later at low fertilizer doses. This may have important implications at the population level because later flowering typically leads to lower seed production (Fried, Kazakou & Gaba, 2012). This is especially significant if seed shedding takes places after the harvest time, which may negatively affect their preservation (Pino, Sans & Masalles, 1998).

Overall, there were no differences between the flowering onset and rarity status. Only the rare *B. testiculata* bloomed significantly later than its paired common species, especially when competing with wheat. A high competition pressure from the crop might have caused this species to become rarer due to reduced reproduction rates over time (Goldberg & Miller, 1990).

Flowering time seemed less sensitive than both biomass and size to the competitive relationship under the different fertilizer regimes. Nevertheless, the limited extent of our sample size must be considered. It could be interesting to collect flowering measurements more often during the peak of the bloom, combined with reproductive allocation of biomass, to obtain a more accurate estimate of the effects of competition on the time to flowering under the different fertilization regimes.

Conclusions

Characteristic arable weed species positively tolerate the application of high doses of both organic and mineral fertilization. However, they suffered under high rates of mineral fertilization due to the indirect effects of cereal competition. Organic fertilization led to a lesser growth of cereal plants compared with the mineral fertilization. This reduction in cereal size allowed characteristic arable weeds to perform better. Flowering was not significantly affected by the competition of wheat at different fertilizer types and doses. Only some characteristic arable species tended to suffer flowering delays under low or organic fertilization regimes, which may involve reduced offspring, affecting the weed community dynamics. Therefore, population reductions for some weeds may stem from resource acquisition effects, more than from changes in the timing of weed development.

Our results suggest that crop competition, which is shaped by the type (organic and mineral) and the dose (low or high) of fertilization, may play a prominent role in contributing to the decline in arable plant diversity. Higher doses of mineral fertilization may have also contributed to the rarity of some currently threatened characteristic species, but this may not be a general pattern. Nevertheless, lower doses of fertilizers or the use of organic fertilization should be promoted to conserve characteristic arable species.

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Supporting information to the paper

ROTCHÉS-RIBALTA, R, BLANCO-MORENO, JM, ARMENGOT, L SANS, FX. Modulation of the competitive relationship between wheat and the characteristic and rare arable species of winter cereal fields by fertilizers. *Weed Research*

Appendix S1: Effects of the fixed factors on the size (mean length of the rosette leaves or total stem length) of characteristic arable plants. Estimated coefficients and their standard errors from the mixed effect models are presented. Significance of the effects is indicated as · when p-value < 0.1, * <0.05, ** < 0.01 and *** < 0.001. Significant differences (p-value < 0.05) in each parameter between the rare (R) and the common (C) species of each family are shaded.

	Bifora testiculata	*Scandix pecten-veneris*	*Neslia paniculata*	*Rapistrum rugosum*
	(Rosette leaves length)	(Rosette leaves length)	(Rosette leaves length)	(Rosette leaves length)
Intercept	3.763 ± 0.091***	4.225 ± 0.093***	3.834 ± 0.144***	3.912 ± 0.223***
Type of fertilizer				
Organic (vs. Mineral)	-0.491 ± 0.118**	-0.405 ± 0.117**	-0.366 ± 0.136*	-0.531 ± 0.280·
Dose of fertilizer				
High fertilizer (vs. Low fertilizer)	0.302 ± 0.118*	0.368 ± 0.116**	0.471 ± 0.136**	0.563 ± 0.280·
Sowing wheat density				
Wheat (vs. No wheat)	-0.509 ± 0.118***	-0.308 ± 0.117*	-0.576 ± 0.136***	-0.520 ± 0.280·
Sampling date				
Sampling date (Linear term)	0.617 ± 0.049***	0.535 ± 0.407***	0.534 ± 0.068***	0.785 ± 0.071***
Sampling date (Quadratic term)	-0.126 ± 0.049*	-0.211 ± 0.040***	-0.171 ± 0.068*	-0.034 ± 0.070
Sampling date (Cubic term)	-0.097 ± 0.049·	-0.035 ± 0.039	-0.032 ± 0.068	0.029 ± 0.072
Organic fertilizer (vs. Mineral) × High fertilizer (vs. Low)	-0.311 ± 0.136*	-0.216 ± 0.135	-0.231 ± 0.157	-0.203 ± 0.323
Organic fertilizer (vs. Mineral) × Wheat (vs. No wheat)	0.186 ± 0.136	0.175 ± 0.135	0.164 ± 0.157	0.068 ± 0.323
High fertilizer (vs. Low) × Wheat (vs. No wheat)	0.259 ± 0.136·	-0.198 ± 0.135	-0.067 ± 0.157	-0.219 ± 0.323
Organic fertilizer (vs. Mineral) × Sampling date L	-0.143 ± 0.049**	0.090 ± 0.042*	0.196 ± 0.068**	0.068 ± 0.068
Organic fertilizer (vs. Mineral) × Sampling date Q	0.082 ± 0.049·	0.053 ± 0.041	0.011 ± 0.068	0.061 ± 0.067
Organic fertilizer (vs. Mineral) × Sampling date C	0.003 ± 0.049	-0.048 ± 0.041	-0.081 ± 0.068	-0.027 ± 0.067
High fertilizer (vs. Low) × Sampling date L	0.074 ± 0.049	-0.038 ± 0.041	0.038 ± 0.068	-0.019 ± 0.068
High fertilizer (vs. Low) × Sampling date Q	-0.052 ± 0.049	-0.030 ± 0.041	-0.125 ± 0.068·	-0.126 ± 0.067·
High fertilizer (vs. Low) × Sampling date C	0.017 ± 0.049	0.066 ± 0.040	-0.032 ± 0.068	-0.112 ± 0.067·
Wheat (vs. No wheat) × Sampling date L	-0.218 ± 0.049***	-0.173 ± 0.042***	-0.049 ± 0.068	-0.213 ± 0.068**
Wheat (vs. No wheat) × Sampling date Q	0.114 ± 0.049*	0.181 ± 0.041***	0.068 ± 0.068	-0.001 ± 0.067
Wheat (vs. No wheat) × Sampling date C	0.024 ± 0.049	-0.066 ± 0.041	-0.018 ± 0.068	-0.108 ± 0.067

Appendix S1 (Continued)

	Papaver argemone (Rosette leaves length)	*Papaver rhoeas* (Rosette leaves length)	*Asperula arvensis* (Total stem length)	*Galium spurium* (Total stem length)
Intercept	2.778 ± 0.238***	3.478 ± 0.234***	4.148 ± 0.387***	5.350 ± 0.273***
Type of fertilizer				
Organic (vs. Mineral)	-0.041 ± 0.310	-0.949 ± 0.306**	-1.752 ± 0.490**	-1.413 ± 0.355**
Dose of fertilizer				
High fertilizer (vs. Low fertilizer)	1.005 ± 0.310**	0.969 ± 0.306**	1.357 ± 0.485*	0.584 ± 0.356
Sowing wheat density				
Wheat (vs. No wheat)	-0.461 ± 0.274	-1.080 ± 0.306**	-1.080 ± 0.447*	-1.628 ± 0.355***
Sampling date				
Sampling date L	0.928 ± 0.064***	1.032 ± 0.072***	2.624 ± 0.152***	2.357 ± 0.119***
Sampling date Q	-0.188 ± 0.063**	-0.105 ± 0.071	-0.076 ± 0.143	-0.140 ± 0.119
Sampling date C	-0.125 ± 0.062*	-0.190 ± 0.071**	-0.463 ± 0.136***	-0.310 ± 0.119*
Organic fertilizer (vs. Mineral) × High fertilizer (vs. Low)	-0.823 ± 0.378·	-0.343 ± 0.353	0.278 ± 0.586	-0.812 ± 0.411·
Organicl fertilizer (vs. Minera) × Wheat (vs. No wheat)	-0.053 ± 0.316	0.756 ± 0.353*	1.662 ± 0.516**	1.042 ± 0.411*
High fertilizer (vs. Low) × Wheat (vs. No wheat)	-0.423 ± 0.316	-0.157 ± 0.353	-1.439 ± 0.516*	-0.110 ± 0.411
Organic fertilizer (vs. Mineral) × Sampling date L	-0.109 ± 0.064·	0.215 ± 0.070**	-0.538 ± 0.154***	-0.257 ± 0.120*
Organic fertilizer (vs. Mineral) × Sampling date Q	0.125 ± 0.063·	0.208 ± 0.070**	-0.045 ± 0.147	0.185 ± 0.120
Organic fertilizer (vs. Mineral) × Sampling date C	-0.053 ± 0.063	-0.141 ± 0.069*	0.136 ± 0.142	-0.084 ± 0.119
High fertilizer (vs. Low) × Sampling date L	0.149 ± 0.064*	-0.013 ± 0.070	0.432 ± 0.156**	0.129 ± 0.120
High fertilizer (vs. Low) × Sampling date Q	-0.090 ± 0.063	-0.187 ± 0.070**	0.047 ± 0.147	0.017 ± 0.120
High fertilizer (vs. Low) × Sampling date C	0.015 ± 0.063	0.096 ± 0.069	-0.043 ± 0.142	-0.114 ± 0.119
Wheat (vs. No wheat) × Sampling date L	-0.382 ± 0.064***	-0.532 ± 0.070***	-1.224 ± 0.157***	-0.893 ± 0.120***
Wheat (vs. No wheat) × Sampling date Q	0.217 ± 0.064***	0.086 ± 0.070	0.183 ± 0.150	0.046 ± 0.120
Wheat (vs. No wheat) × Sampling date C	-0.006 ± 0.063	-0.011 ± 0.069	0.581 ± 0.145***	0.179 ± 0.119