



HAL
open science

Analysis of the agronomic and economic performances of lentil-spring wheat intercrops in organic farming

Loïc Arthur Viguiier

► To cite this version:

Loïc Arthur Viguiier. Analysis of the agronomic and economic performances of lentil-spring wheat intercrops in organic farming. Agricultural sciences. Institut National Polytechnique de Toulouse - INPT, 2018. English. NNT : 2018INPT0070 . tel-04227235

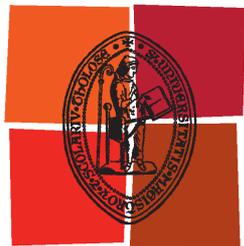
HAL Id: tel-04227235

<https://theses.hal.science/tel-04227235>

Submitted on 3 Oct 2023

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.



Université
de Toulouse

THÈSE

En vue de l'obtention du

DOCTORAT DE L'UNIVERSITÉ DE TOULOUSE

Délivré par :

Institut National Polytechnique de Toulouse (Toulouse INP)

Discipline ou spécialité :

Agrosystèmes, Ecosystèmes et Environnement

Présentée et soutenue par :

M. LOIC VIGUIER

le jeudi 12 juillet 2018

Titre :

Analyse de la performance agronomique et économique des associations de culture lentille-blé de printemps en agriculture biologique

École doctorale :

Sciences écologiques, vétérinaires, agronomiques et bioingénierie (SEVAB)

Unité de recherche :

AGroécologie, Innovations, TeRritoires (AGIR)

Directeur(s) de Thèse :

M. ETIENNE-PASCAL JOURNET

M. ERIC JUSTES

Rapporteurs :

M. GEORG CARLSSON, ALNARP

M. AGNES PIQUET-PISSALOUX, VERTAGROSUP

Membre(s) du jury :

MME ALEXANDRA JULLIEN, AgroParisTech, Présidente

M. ETIENNE-PARIS JOURNET, CNRS-INRA, Membre

M. LAURENT BEDOUSSAC, UNIVERSITE DE TOULOUSE, Membre

MME MURIEL VALANTIN-MORISON, AgroParisTech, Membre

Thesis manuscript

“Analysis of the agronomic and economic performances of lentil-spring wheat intercrops in organic farming”

Loïc Viguié

Thesis supervisors:

Laurent BEDOUSSAC, Maître de Conférences ENSFEA, Co-Encadrant de thèse

Etienne-Pascal JOURNET, Chargé de recherche CNRS, Directeur de thèse

Eric JUSTES, Cadre Supérieur CIRAD, Co-Directeur de thèse

UMR AGIR "Agro-écologie - Innovations - Territoires", Centre INRA de Toulouse-Auzeville

Ecole Doctorale SEVAB, Institut Polytechnique National de Toulouse

Thèse CIFRE (ANRT et coopérative QUALISOL)

Participant to the defense as members of the jury:

Georg CARLSSON, Senior Lecturer SLU, Alnarp, Suède, Rapporteur

Agnès PIQUET-PISSALOUX, Professeure VetAgroSup, Rapportrice

Muriel VALANTIN-MORISON, Directrice de recherche INRA, AgroParisTech, Examinatrice

Alexandra JULLIEN, Professeure AgroParisTech, Examinatrice

Laurent BEDOUSSAC, Maître de Conférences ENSFEA, Co-Encadrant de thèse

Etienne-Pascal JOURNET, Chargé de recherche CNRS, Directeur de thèse

Participant to the defense as guests:

Eric JUSTES, Cadre Supérieur CIRAD, Co-Directeur de thèse

Alain Larribeau, Directeur pôle agronomique coopérative QUALISOL, Co-Encadrant de thèse

Remerciements

Je tiens à remercier, de manière générale, toutes les personnes qui m'ont permis de réaliser ce travail.

Plus précisément, je remercie mes trois encadrants : Laurent Bedoussac, Etienne-Pascal Journet et Eric Justes. Merci pour votre exigence qui m'a permis d'évoluer et merci d'avoir été présents tout au long de ces trois années. Je suis heureux d'avoir réalisé ma thèse sous votre direction et vos enseignements me serviront tout au long de ma vie professionnelle. Je remercie aussi vivement Alain Larribeau, directeur du pôle agronomique de la coopérative Qualisol, de m'avoir fait confiance et d'avoir toujours témoigné de l'intérêt vis-à-vis de mes travaux. Les restitutions aux agriculteurs ont été pour moi une très bonne expérience.

Je remercie également tous les membres de mon jury : Alexandra Jullien (présidente), Georg Carlsson (Rapporteur), Agnès Piquet-Pissaloux (Rapporteur) et Muriel Valentin-Morison (examinatrice), d'avoir évalué mon travail. Vos commentaires amélioreront grandement les articles à soumettre.

Je remercie aussi les membres de mon comité de thèse : Pierre Casadebaig, Gaëtan Louarn, Delphine Moreau, Christophe Naudin pour leurs remarques pertinentes qui ont permis de consolider mes hypothèses et mes protocoles expérimentaux.

Je remercie Dominique Pantalacci et Claudes Maranges de l'école doctorale SEVAB.

Je remercie Issam Moussa de la plateforme isotopique d'ECOLAB, Olivier Delfosse de l'INRA de Reims et la plateforme PLATIN, pour les analyses élémentaires.

Je remercie chaleureusement Jérémy Grève (coopérative Qualisol) d'avoir été présent dès que j'en avais besoin. Ce fut un réel plaisir de travailler avec toi, je te souhaite beaucoup de réussite et de bonheur pour la suite.

Je remercie toutes les personnes de l'UMR AGIR et notamment Jacques-Eric Bergez (directeur), pour la bonne ambiance et les échanges. J'ai beaucoup apprécié travailler avec vous durant ces trois années. Je remercie aussi Sandra Fuentes de la communication, notamment pour avoir pensé à nous pour les ateliers avec les enfants.

Je remercie tous les techniciens de l'UMR AGIR et du domaine expérimental et plus particulièrement Benoît Gleizes, Didier Chesneau, Eric Lecloux et Didier Raffailac. Je

remercie aussi Michel Labarrère qui m'a beaucoup aidé tout au long de mes expérimentations et sans qui je n'aurais pas pu réaliser ce travail.

Je remercie vivement Christel Model et Maryse Perreu de l'UMR AGIR et j'ai une pensée particulière pour Marina Lefebvre que j'ai bien connue pendant ces trois ans.

Je remercie tous les stagiaires et les aides occasionnelles que j'ai pu rencontrer pendant ces trois ans. Merci à Estelle Teyssier d'avoir réalisé son stage sur mon projet, j'espère que tu trouveras une thèse qui te plaira !

Je remercie également tous les doctorants d'AGIR et bien sûr le bureau 205 : Céline Schoving, Sixtine Cueff (partie trop tôt), Nicolas Meyer et Antoine Couëdel. J'espère retrouver une telle ambiance dans mon prochain boulot. Sans vous, ces trois années n'auraient pas eu la même saveur ! Merci beaucoup et à votre tour maintenant ! Pensées également pour Yolaine Hily, Julie Bréchemier et Elise Audoin de l'unité AGIR !

Je remercie toute ma famille pour leur soutien tout au long de ma thèse, ça y est, c'est terminé ! Je remercie aussi tous mes colocs du Kennedy, ceux qui sont partis et ceux avec qui je suis encore. C'était trois belles années et il en reste encore plein à vivre ensemble. Je pense aussi à tous mes amis et ceux de toujours, Bastien et Louis. Enfin je pense à Marjorie, merci d'avoir été au plus près durant les moments difficiles, sans toi ça aurait été beaucoup plus dur !

List of contents

CHAPTER I. GENERAL INTRODUCTION	9
1.1. Adverse effects of the current dominant western agricultural system	9
1.2. Why are grain legumes a relevant lever to increase agriculture sustainability?	12
1.3. Current lock-ins of grain legumes cultivation and use in Europe.....	13
1.4. Why should we be interested in lentils?	14
1.4.1. History and description	14
1.4.2. Major production and consumption areas	15
1.4.3. Nutritional stakes of lentils	15
1.5. Lentil agronomy	16
1.5.1. Crop management	16
1.5.1.1. Soil and place in rotations	16
1.5.1.2. Fertilization	17
1.5.1.3. Cultivars, sowing densities and sowing	17
1.5.2. Major agronomic constraints	18
1.5.2.1. Water and temperature	18
1.5.2.2. Bruchids	18
1.5.2.3. Lodging	18
1.5.2.4. Weeds	19
1.6. Intercropping, an agronomic solution to lentil cropping constraints?	19
1.6.1. Definition and uses	19
1.6.2. General principles of intercropping	20
1.6.3. Knowledge on lentil in intercrops	20
1.6.4. Participation of the agricultural cooperative Qualisol in the thesis	21
1.7. Objectives of the thesis and main research questions	22
1.7.1. Main objectives, research questions and experimental strategy	22
1.7.2. Main research questions	22
1.8. Outline of the thesis	23
 CHAPTER II. YIELD GAP ANALYSIS EXTENDED TO MARKETABLE GRAIN REVEALS THE PROFITABILITY OF ORGANIC LENTIL-SPRING WHEAT INTERCROPS	 24

2.1.	Introduction	26
2.2.	Materials and methods	29
2.2.1.	Improving the yield gap concept.....	29
2.2.2.	Experimental design	31
2.2.3.	Measurements, calculations and statistics	31
2.2.3.1.	Actual yield measured by hand-harvest	31
2.2.3.2.	Mechanical harvest efficiency measurement.....	32
2.2.3.3.	Lentil height, stem length, lodging and lowest pod height.....	32
2.2.3.4.	Estimation of bruchid-damaged grain rate, total grain loss due to bruchids and attainable yield	33
2.2.3.5.	Economic parameters.....	34
2.2.3.6.	Statistical analysis	35
2.3.	Results and discussion.....	35
2.3.1.	Effect of intercrops on actual yield	35
2.3.2.	Effect of intercropping on gross margin from actual yield.....	38
2.3.3.	Effects of intercrops on bruchid damage, lodging and mechanical harvest efficiency	38
2.3.3.1.	Effect of intercrops on bruchid damage	38
2.3.3.2.	Effect of intercrops on lentil height at harvest, stem length and lowest pod height	39
2.3.3.3.	Effect of intercrops on mechanical harvest efficiency.....	41
2.3.4.	Effect of intercrop gross margin from marketable yield	42
2.3.5.	Yield gap analysis of all cultivars and years combined.....	43
2.4.	Conclusion.....	45

CHAPTER III. SPECIES NICHE COMPLEMENTARITY FOR NITROGEN IMPROVES LENTIL-WHEAT INTERCROP GRAIN PRODUCTION IN ORGANIC FARMING..... 46

3.1.	Introduction	48
3.2.	Materials and Methods	50
3.2.1.	Site, soil, and climate	50
3.2.2.	Experimental design	50
3.2.3.	Sampling and measurements	51
3.2.4.	Estimate of attainable lentil yield.....	52
3.2.5.	Calculations and analysis	52
3.2.5.1.	Nitrogen acquisition	52
3.2.5.2.	Nitrogen fixation.....	53
3.2.5.3.	Land equivalent ratio.....	53
3.2.6.	Statistics	54

3.3.	Results	55
3.3.1.	Grain yield and nitrogen acquisition in total intercrops and mean sole crops.....	55
3.3.2.	Species density drives competition in intercrops.....	56
3.3.3.	Land equivalent ratios for grain yield and nitrogen accumulated in shoots.....	57
3.3.4.	Grain yield is strongly correlated with nitrogen accumulated in shoots.....	58
3.3.5.	Lentil fixes a higher percentage of nitrogen in intercrops but in lower amounts	59
3.3.6.	Protein concentration is correlated with grain yield for wheat but not for lentil	60
3.4.	Discussion.....	61
3.4.1.	Grain yield and nitrogen accumulated in the total intercrop vs. sole crops.....	61
3.4.2.	Grain yield of and nitrogen accumulated by intercropped species	62
3.4.3.	Grain protein concentrations of species in intercrops and sole crops	63
3.4.4.	Strategies to improve lentil production in intercrops	64
3.5.	Conclusions	65

CHAPTER IV. YIELD COMPONENT ANALYSIS REVEALS THAT LENTIL-SPRING WHEAT INTERCROP GRAIN YIELD IS DETERMINED EARLY DURING THE VEGETATIVE STAGES..... 67

4.1.	Introduction	69
4.2.	Material and methods.....	71
4.2.1.	Site, soil, and climate	71
4.2.2.	Cultivars and densities	71
4.2.3.	Experimental design and crop sampling	72
4.2.4.	Yield components denomination.....	72
4.2.5.	Yield components measurement and calculation (Fig. 13).....	73
4.2.5.1.	Wheat yield components measurement and calculation	73
4.2.5.2.	Lentil yield components measurement and calculation	74
4.2.6.	Calculation of Interspecific interaction indices for yield components.....	75
4.2.7.	Statistics.....	76
4.3.	Results	77
4.3.1.	Lentil and spring wheat yields	77
4.3.2.	Effect of the year on lentil and wheat yield components.....	79
4.3.3.	Effect of wheat density on lentil and wheat yield components.....	82
4.3.4.	Effect of lentil cultivar on lentil and wheat yield components	84
4.4.	Discussion.....	85
4.4.1.	Global intercrop performances.....	85
4.4.2.	Early impacts of interspecific interactions on species yield components	86

4.4.3.	Intensity of interspecific interactions is affected by year	88
4.4.4.	Wheat density strongly affects lentil yield in intercrops	89
4.4.5.	Sensitivity to interspecific competition varies among genotypes, especially in lentil	90
4.5.	Conclusion	91
 CHAPTER V. SYNTHESIS, GENERAL DISCUSSION AND PERSPECTIVES		93
5.1.	Synthesis of main results	93
5.1.1.	Higher nitrogen acquisition and total grain yield of intercrops	93
5.1.2.	Wheat intercrop grain protein concentration is increased	94
5.1.3.	Interspecific competition began early in vegetative growth	95
5.1.4.	Intensity of interactions between plants depends on climatic conditions, densities and cultivars 96	
5.1.4.1.	Effect of climatic conditions	96
5.1.4.2.	Effect of density	97
5.1.4.3.	Effect of genotype	98
5.1.5.	Effect of intercrops on lentil agronomic constraints	98
5.1.5.1.	No observed effect of intercrops on bruchid damages	98
5.1.5.2.	Reduction of late emergence weeds in intercrops	99
5.1.5.3.	Reduction of lentil lodging by intercrop wheat leads to a better lentil harvest ability and a higher intercrop profitability	100
5.2.	General conclusion	102
5.3.	Perspectives	104
5.3.1.	Which management of intercrops at the Qualisol cooperative level	104
5.3.2.	Propositions for further research	105
5.3.2.1.	Research on bruchids is needed	105
5.3.2.2.	Further knowledge on lentil eco-physiology in relation to cultivar diversity is also needed 106	
 BIBLIOGRAPHY		108
 LIST OF FIGURES		118
 LIST OF TABLES		120
 ABSTRACT		121

General introduction

1.1. Adverse effects of the current dominant western agricultural system

Conventional farming is the historical model of industrial agriculture in Western countries (Plumecocq et al. 2018). It is characterized by a high use of synthetic inputs and a field crop specialization (Magrini et al. 2016). Conventional farming development was triggered by the discovery of the Haber-Bosch process that synthesizes ammonia (a precursor of useable mineral forms of nitrogen for plants) from atmospheric dinitrogen (N_2), at the beginning of the 20th century (Smil 2002; Erisman et al. 2008). The conventional model of agriculture has achieved its objective of producing high, affordable, amounts of food as shown by an increase in the number of humans supported per hectare of arable land from 1.9 to 4.3 persons between 1908 and 2008 on a world scale (Erisman et al. 2008). Along with the use of synthetic pesticides, the high use of nitrogen (N) fertilizers explains to a large extent the increase in land productivity. At the onset of the 21th century, 44% of the world's population was fed thanks to the use of nitrogen fertilizers in agriculture (Smil 2002; Erisman et al. 2008). As an example of the success of conventional agriculture in producing high amounts of food, Europe (28 states) is at the beginning of the 21st century self-sufficient in key agricultural commodities such as major meats (e.g. pigmeat, poultry, beef), milk and cereals and is even a net exporter in these commodities (Watson et al. 2017).

Yet, the real cost of achieving food security through conventional farming is higher than the current price of staples. The adverse impacts (also referred to negative externalities) on the environment of such a model have been reported by a large body of science (German et al. 2016). The greenhouse gas (GHG) emissions of the worldwide food chain, that includes preproduction (input fabrication), production (direct and indirect emissions from agriculture) and postproduction (transport, processing) account for 19%-29% of total global anthropogenic GHG emissions and within that food chain, production contributes to 80-86% of the emissions (Vermeulen et al. 2012; IPCC 2014). The anthropogenic cause to climate change is now considered as “extremely likely” (IPCC 2014) and climate change is recognized as “one of the greatest challenges to food security” especially through increased climate variability leading to yield instability (Vermeulen et al. 2012). Actually, human activities have exerted such an impact on the planet's environment that the term “Anthropocene” has been proposed to distinguish the last part of the current geological epoch, the “Holocene” (Crutzen 2006;

Rockström 2009). In addition to GHG emissions, industrial agriculture through its intensive land use also participates to chemical pollution (Rockström 2009), soil degradation (German et al. 2016; Cellier 2017) and biodiversity loss (Laurance et al. 2014).

It is obvious that the continuation of conventional agriculture in a “business as usual” way threatens the global food system’s sustainability, defined by the Bruntland report (World Commission on Environment and Development 1987) as “the development that meets the needs of the present without compromising the ability of the future generations to meet their own needs”. Nowadays, the dominant form of agriculture in Europe is called “technology-intensive” (Müller and Kreuer 2016). This model is very close to the historical conventional one, but it includes the use of innovative technologies such as genetic engineering and precision farming in order to increase input utilization efficiency (Plumecocq et al. 2018). Indeed, if we take the example of N application in the western world, half of the total amount applied in agriculture is currently lost to the environment generating air and water pollution (Cellier 2017). Many authors agree that one step towards higher sustainability is to increase the use efficiency of inputs (Erisman et al. 2008; Jensen et al. 2011).

Therond et al. (2017) suggested two axes of characterization of agriculture models: (1) exogenous inputs versus ecosystem services and (2) low versus high territorial embeddedness. Plumecocq et al. (2018) proposed six different sustainable agriculture models using Therond’s typology (Fig. 1).

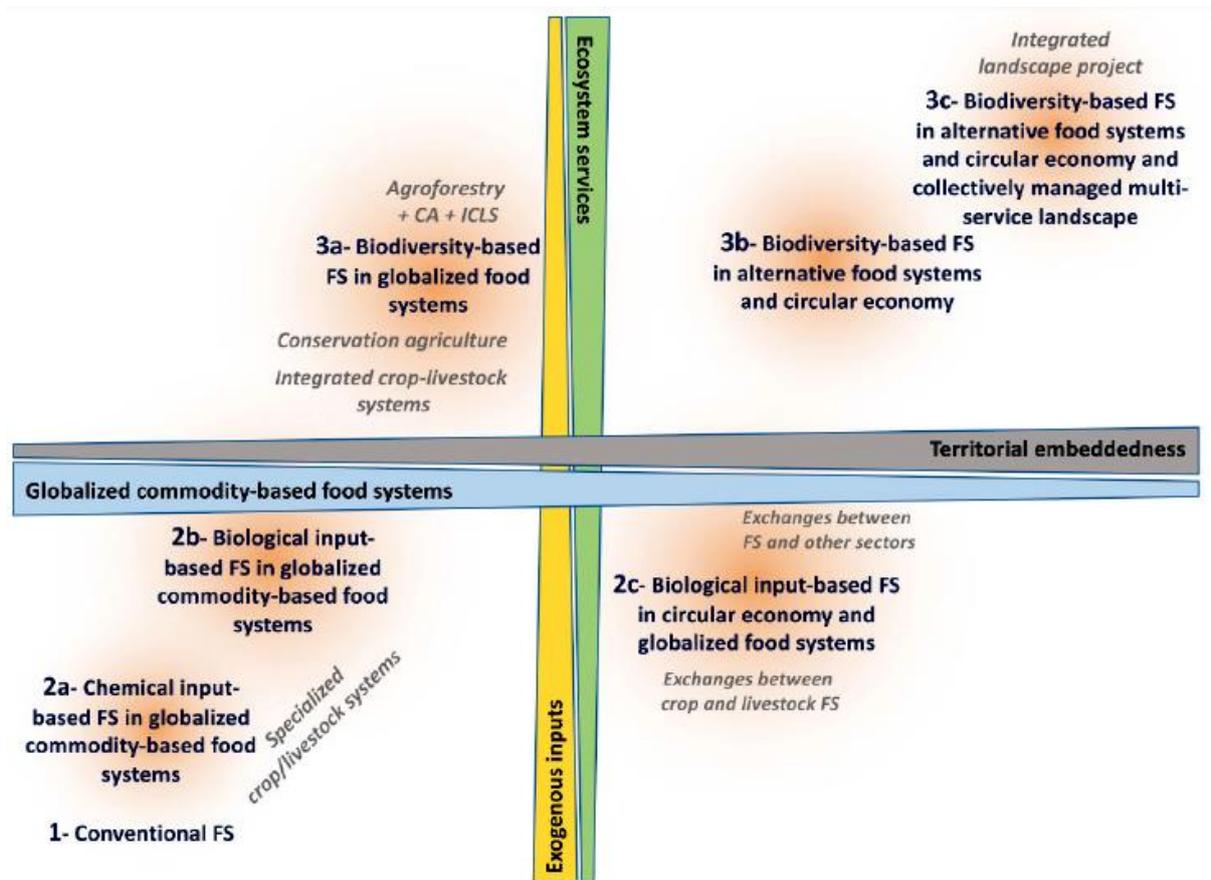


Fig. 1 Main models of agriculture (from 1 to 3b in blue) with farming systems identified based on their varying degrees of use of ecosystem services versus anthropogenic exogenous inputs (Y-axis) and connected to globalized food systems or local dynamics (X-axis). Iconic examples are presented in grey. The number 1 is for conventional farming systems outside less-favored areas (1 being the current, conventional agriculture model). The main alternative agriculture models were grouped into two types of alternatives to reflect the paradigm shift between input-based (type-2) versus biodiversity-based farming systems (type-3). Submodels labeled a, b, and c mainly reflect the relationships between farming systems, globalized food systems, and local dynamics. (CA: conservation agriculture; FS: farming system; ICLS: integrated crop livestock systems) (from Plumecocq et al. 2018).

For instance, the dominant “technology-intensive” model (2a in Fig. 1) was defined by high exogenous inputs (e.g. N inputs) and low territorial embeddedness (i.e. food production is oriented towards the global market). The objective of this thesis is not to discuss the different forms of sustainable agriculture but, interestingly, Therond’s typology shows that the alternative of high exogenous inputs is the use of ecosystem services defined as service that “ecosystems provide to humanity such as soil formation, nutrient cycling, water supply, pollination and biological control of pests“, (Iverson et al. 2014). Among these numerous ecosystem services, the N fixation ability of legume plants is of particular interest since it can help to reduce N inputs and hence GHG emissions from agriculture (Schneider et al. 2015; Watson et al. 2017). Organic agriculture can contribute to increasing sustainability of farming

systems because use of synthetic inputs is prohibited. This model of agriculture is based on a more efficient use of ecosystem services, particularly biological nitrogen fixation (Jensen et al. 2003) via the incorporation of legumes in rotations (Seufert et al. 2012).

1.2. Why are grain legumes a relevant lever to increase agriculture sustainability?

Grain legumes are annual plants of the Fabaceae family harvested for their grain (Nguyen 2018). They include inter alia and per order of total production worldwide in 2011: soybean (*Glycine max* L.), groundnut (*Arachis hypogaea* L.), bean (e.g. *Phaseolus vulgaris* L.), chickpea (*Cicer arietinum* L.), pea (*Pisum sativum* L), faba bean (*Vicia faba* L.) and lentil (*Lens culinaris*; Duc et al. 2015). They represent the second most economically important family of agricultural crop species after grasses (i.e. Poaceae, such as corn or wheat; Watson et al. 2017; Willis 2017). Note that the commonly used term “pulses” refers only to grain legumes that are harvested for their dry grain, so that soybean and groundnut are not included in that denomination because of the high oil content of their seeds and are often referred as “Oilseed” (Duc et al. 2015). Legumes are known primarily for their ability to transform atmospheric N₂ into ammonium (NH₄⁺) a plant metabolisable form of N, through a process called “Biological Nitrogen Fixation, (Voisin and Gastal 2015). Biological N fixation is the result of a symbiotic relationship between the legume and N-fixing rhizobacteria that trigger on the legume roots the development of specific organs, the so-called nodules, where rhizobacteria are accommodated. The rhizobacteria fix N₂ in exchange for nutrients from the legume (Voisin and Gastal 2015; Nguyen 2018). Before the development of N fertilizers, nearly all the reactive N introduced into farming systems was from biological N fixation (Cellier 2017). At the dawn of the 21st century, anthropogenic reactive N production has overtaken that of legumes on a global scale (Cellier 2017) contributing, as mentioned earlier, to the increase of global food production but at the expense of pollution: e.g. carbon dioxide (CO₂) emissions associated with fertilizer conception and use (Cellier 2017) or surplus of nitrous oxide (N₂O) emissions through nitrification and denitrification of excessive N inputs (Jeuffroy et al. 2013).

The agronomic advantages of legumes depend to a large extent on the biological N fixation. Many legumes do not require N fertilization for grain production because they can meet most of their N needs through BNF (Peoples et al. 2009). Thus, incorporating legumes into crop rotations reduces per se the amount of N application and GHG emissions related to fertilizer

use. Jeuffroy et al. 2013 showed that pea generated 4-6 times less N₂O emissions per unit area in France than fertilized wheat.

Legumes preferentially use soil mineral nitrogen since the total energy cost of N acquisition from fixation is higher than that of soil nitrate assimilation (NO₃⁻; Voisin and Gastal 2015). Consequently, the percentage of N fixed from the air by legumes (%Ndfa) is negatively correlated to mineral N, (Voisin and Gastal 2015) and the optimum level of N_{min} for high %Ndfa is below 50-56 kg N ha⁻¹ (Voisin et al. 2002). Consequently, to maximize the biological nitrogen fixation, legumes should be introduced in soils with low nitrogen content.

Grain legumes can fix on average 100-200 kg N ha⁻¹ yr⁻¹ in their above-ground biomass (Nguyen 2018) and part of the N non allocated to grains can be incorporated into the soil through exudates and biomass residues to provide N to the following crops (Peoples et al. 2009; Preissel et al. 2015). This “pre-crop effect” can increase yields of about 0.5-1.6 t ha⁻¹ for a following cereal (Zander et al. 2016; Preissel et al. 2015). Incorporation of residues also contributes improving the soil organic carbon content hence soil structure hence nutrient and water holding capacity (Nguyen 2018). Finally, cultivating legumes improves in-field biodiversity and cropping management variation, which helps break pathogen and weed cycles and reduce pesticide use (Peoples et al. 2009; Angus et al. 2015).

Despite the many agronomic advantages of legumes that can help lower agriculture pollution and mitigate GHG, they are currently underrepresented on a global scale, and especially in Europe. The main reasons for this situation will be presented in the following section.

1.3.Current lock-ins of grain legumes cultivation and use in Europe

Grain legumes are hardly used by farmers in Europe where they represented only 1.5% of the arable area in 2014 (14.5% at global scale, Watson et al. 2017). This is a result of a long trend which started with the development of conventional agriculture. From the shortening of crop rotations emerged highly organized supply chains dedicated to “major” crops (mainly cereals such as wheat and maize) that obliterated “minor” crops, including grain legumes (Magrini et al. 2016). Stakeholders (researchers, advisors, farmers, cooperatives and agro-industry) developed their activities towards these major crops so that their combined efforts gradually improved the performance of these crops within the conventional system (Arthur 1994; Magrini et al. 2016). This is illustrated by the linear increase in cereal yields within the EU from 1961 to 2016 (57 kg ha⁻¹ yr⁻¹; World Bank 2018) even if signs of yield stagnation have appeared since 2000 in high producing countries such as France (Gate and Gouache 2010). Consequently,

minor crops that have not benefited for important genetic improvement, agronomic research, experience from farmers and market development are perceived as risky by stakeholders who turn away from them. The lock-in of grain legumes in Europe has thus been strengthening with time, with the technical and economic improvement of cereal performance (Magrini et al. 2016). This concept of lock-in has been theorized by Arthur (1994) and is called Increasing Returns of Adoption.

Nevertheless, the EU seems willing to begin unlocking the grain legume situation as shown by this recently adopted text at the European Parliament (April 2018) stressing the importance of developing legumes within the Union: “ It is vital to reduce the Union’s massive dependency on imports of protein crops [...], vegetable proteins are at the core of the challenges of food security and sovereignty, environmental protection, global warming and renewable energy [...], the European parliament recommends supporting, in particular under the Common Agricultural Policy, the cultivation of soya in the EU by making it profitable and competitive [...], but (EU) notes that this should not overshadow the cultivation of other grain protein crops (European strategy for the promotion of protein crop, 2018).

We will now focus on the grain legume lentil that has an interesting role to play within this context of grain legume development in Europe that faces multiple economic and also, as we will see in the next sections, agronomic difficulties.

1.4. Why should we be interested in lentils?

1.4.1. History and description

Lentil (*Lens culinaris* Medikus subsp. *Culinaris*) was one the first crops domesticated by Man in the Fertile Crescent during the Neolithic period, i.e. around 10000 BC (Sandhu and Singh, 2007). The genus *Lens* comprises six species and only *L. culinaris* Medikus is used for agriculture. *L. culinaris* is divided into two sub-species differing in the size of their seeds (1) *microsperma* (seed diameter, 2-6 mm) and (2) *macrosperma* (seed diameter, 6-9 mm; Redden et al. 2007). Lentil is a bushy herb with semi-erect, spreading and compact growth (Erskine et al. 2009). Lentil stems are slender and about 15-75 cm in height (Sandhu and Singh, 2007). Lentil is self-pollinating, indeterminate plant with flowering occurring acropetally (Andrews and McKenzie 2007; Sandhu and Singh, 2007). Axillary racemes carry 1-4 flowers on short peduncles and pods generally contain 1-3 seeds whose shape, size, colour and nutritional content vary with cultivars. The International Center for Agricultural Research in Dry Areas

(ICARDA) is the worldwide reference on lentil accession conservation. About half of the collection is made up of accessions from Central and West Asia and North Africa where lentil originated and where it remains an important crop today (Redden et al. 2007).

1.4.2. Major production and consumption areas

Lentil seeds are eminent components of human diets in populated areas of the world such as Asia or Africa where they constitute a substitute to meat proteins mainly because of their affordability and their relative productivity on dry, marginal lands (Savage 1991; Stefaniak and McPhee 2015). About 50 countries produce lentil worldwide. The top producing countries in 2013 were Canada (42% of the world's production) and India (21%; FAOSTATS, 2018). The world production of lentil has been increasing exponentially since 1960 with a 6-fold increase until 2016 (FAOSTATS, 2018). The worldwide average lentil grain yield was 1.2 t ha⁻¹ in 2016, which is slightly higher than the European average (1.0 t ha⁻¹; FAOSTATS, 2018). On a world scale, about two thirds of the lentil production is consumed where it is produced, especially in Asia and Africa (Stefaniak and McPhee 2015). India and Bangladesh are the main importers, confirming the importance of this crop in southern Asia (FAOSTATS, 2018).

EU lentil consumption (EU Production + Import – Export) has steadily increased since 1960, though in 2013 EU produced only 26% of its consumption (FAOSTATS, 2018), and France 45%. This situation can be generalized to grain legumes and is mainly due to the lock-ins discussed earlier. Yet, lentil consumption is likely to further increase in the coming years with (1) increased communication about the health benefits of grain legumes (e.g. FAO 2016 International Year of Pulses) and (2) the political will of the EU to develop these types of crops (European strategy for the promotion of protein crop, 2018). As a sign of the transition towards higher use of grain legumes in Europe, agro-food industries have also started to develop strategies to introduce food product innovations with grain legumes such as bread or “steaks” made of legumes (Lascialfari and Magrini 2016).

1.4.3. Nutritional stakes of lentils

The nutritional advantages of lentil are also an important stake in the development of grain legumes. The adverse impacts of the current western food system have contributed to the increase of heart diseases, type two diabetes and obesity (Polak et al. 2015; Duru et al. 2017). As a response to this situation, the use of plant-based diets is recommended to limit the risk of the occurrence of these diseases (Polak et al. 2015; Duru et al. 2017). A plant-based food system relies to a great extent on the consumption of grain legumes, including lentils, to cover protein

requirements. Lentil has many nutritional advantages with a high protein content (20-36%; Savage 1991), low carbohydrate and fat content and when associated with a cereal, they constitute an amino-acid-balanced diet (Chardigny and Walrand 2016). Furthermore, lentil has a relatively high ash content and is a source of micronutrients such as iron or zinc and vitamins such as folate (B9; Faris et al. 2012). Lentil can be promoted to improve public health, as for other grain legumes in general. This was the main message of the FAO 2016 International Year of Pulses because, unfortunately, these nutritional advantages were generally not well known to the public but are the key to a greater consumption of grain legumes.

Given the nutritional advantages of lentil, the will of the EU to develop grain legumes and the current deficit in lentil production, it seems necessary to increase lentil production in Europe and especially the organic lentil that also has a significant market demand. However, some important agronomical locks exist and currently limit its production in Europe.

1.5.Lentil agronomy

1.5.1. Crop management

1.5.1.1. Soil and place in rotations

Lentil can be grown on a large range of soil types and pH (Andrews and McKenzie 2007; Sekhon et al. 2007) but it is sensitive to waterlogged and saline soils that inhibit BNF (Quinn 2009).

In Europe, lentil is generally grown in sequential cropping i.e. growing two or more crops in a sequence, planting the succeeding crop after the harvesting of the previous one (Wang 2012). The time between two lentil sowings on a same field should not be less than three years to lower the risk of the occurrence of several important diseases such as *Ascochyta lentil*, a fungus causing ascochyta blight that reduces grain yield and quality (Wang 2012). The use of pesticides to control biotic reducing factors (fungi, insects and weeds) is frequent in conventional lentil partially due to short rotations.

Lentil is generally placed before a winter cereal in crop rotations so that the cereal can benefit from N fixed by the lentil. Note that the average amount of N fixed by lentil is relatively low because of its low biomass production compared to that of other grain legumes (71 kg N ha⁻¹ for lentil versus 137 kg N ha⁻¹ for soybean and 108 kg N ha⁻¹ for pea; Peoples et al. 2002; Quinn 2009). Lentil generally have low harvest index (ratio of harvested grain over the total shoot dry matter) compared to other legumes and cereals (ca. 0.40; Whitehead et al. 2000; Solanki et al.

2007; Erskine et al. 2009) so that lentil straw can be valuable for animal feed in integrated crop livestock systems or for soil incorporation (Erskine et al. 1990).

1.5.1.2. Fertilization

Fertilization on lentil is not frequent. Lentils tend to have a high %Ndfa compared to other grain legumes (74% for lentil versus 62% for soybean or 70% for pea) so that they compensate quite efficiently low mineral N availability by BNF (Peoples et al. 2002; Ali et al. 2009). N fertilisation has not been reported to substantially increase lentil yields (McNeil and Materne, 2007) although an addition of small amounts of N (10-25 kg N ha⁻¹) in soils with very low N availability has been shown to promote early growth and BNF (McKenzie et al. 2007). For other macronutrients, application of phosphorus (P), potassium (K) or sulphur (S), can improve lentil yields (Yadav et al. 2009) and, to a lesser extent, applications of essential micronutrients such as zinc (Zn), manganese (Mn), iron (Fe), molybdene (Mo) and bore (B; Yadav et al. 2009).

1.5.1.3. Cultivars, sowing densities and sowing

The choice of a cultivar is particularly important for lentil because genotype × environment interactions are usually significant (Yadav et al. 2009). However, market preferences often force farmers to grow particular cultivars such as the green lentil “Anicia” for the “lentille verte du Puy” which limits the possibility to adapt the lentil cultivar to the prevailing climate.

The effect of plant density on lentil yield is variable and depending upon genotype, date of sowing and environment (Yadav et al. 2009). In Canada, the recommended plant population varies from 107 to 215 plants m⁻² (Muehlbauer et al. 1998) and is higher for organic lentil production (ca. 300 plants per m²; Baird 2009).

Lentil is mainly grown as a spring crop in Europe and early sowings, i.e. starting from end of February, are recommended to lower risks of exposure to drought during the late reproductive cycle in July (Kigel et al. 2015). Soil is usually rolled after sowing to improve seed-soil contact and obtain fast, homogeneous seed emergence (Diekmann and Al-Saleh, 2009). It is also a way to facilitate crop harvest which requires a low height cutting bar because of the lodging habit of lentil (Diekmann and Al-Saleh, 2009).

1.5.2. Major agronomic constraints

1.5.2.1. Water and temperature

Water deficit during vegetative growth can reduce the accumulation of aerial biomass at maturity up to 32-61% (Ashraf et al. 1992; Turay et al. 1992; Shrestha et al. 2006). Water stress lowers the number of leaves, the leaf area and hastens leaf senescence thus directly affecting the photosynthesis of lentil (Shrestha et al. 2009). Lack of sufficient water in the post-flowering period, i.e. during pod filling and maturing, affects the reproductive structures of the plant and leads to a reduced number of filled pods (Rahman et al. 2009). Water stress is amplified by the fact that lentils are generally rainfed in Europe (Ali et al. 2009).

Responses to water deficits differ according to lentil genotypes and some cultivars may benefit from a mild, short, water stress during early flowering which may increase flower production, harvest index and final grain yield (Shrestha et al. 2009). Cool temperatures during vegetative growth lower biomass development and high temperatures in summer increase evapotranspiration, contributing to the occurrence of water stress (Shrestha et al. 2009).

1.5.2.2. Bruchids

Bruchid beetles (Coleoptera: Bruchidae) may decrease grain yields greatly, particularly in organic farming. Grain damaged by bruchids is not marketable as food and represents net yield and income losses for farmers that can exceed 50% (Laserna-Ruiz et al. 2012). Two established bruchid species (*Bruchus lentis* and *B. signaticornis*) can damage lentil in Europe (Delobel 2005; Yus-Ramos et al. 2014). Adults lay eggs on the surface of developing pods, and larvae then penetrate the pod and feed on the growing lentil grain. These two species can damage lentil only in the field, as they are univoltine and do not lay eggs on stored grain (Yus-Ramos et al. 2014). Lentil genotypes tolerant to bruchids have not been reported (Laserna-Ruiz et al. 2012) and no effective biocontrol method is currently available in the field for organic farmers, which hinders development of lentil in areas where bruchid damage is high. Note that synthetic insecticides are available for conventional farming but are not efficient since sufficient crop protection requires many spreadings.

1.5.2.3. Lodging

Lodging is another common issue for lentil which results from a combination of genotypic and environmental factors (Ball et al. 2006). Lentil shoots spontaneously tend to lean to the ground upon maturation and collapse if unfavourable weather (e.g. rain and wind) occurs before harvest

(Sidahmed and Jaber 2004). Impacts of lodging on grain yields may be limited in southern and western Asia, where most lentil is harvested by hand. However, in areas where lentil harvest is mechanical, such as North America or Europe, combine harvesters may fail to pick up plants leaning too much toward the ground, leading to large grain loss in the field, sometimes up to 100% (Carr et al. 1995). Factors besides lodging influencing grain loss during mechanical harvest are numerous, such as weather conditions, field topography, ground roughness, management practices, crop maturity, type of combine harvester used and its settings and height of cut (Erskine and Goodrich 1988; Ibrahim et al. 1993). Erskine and Goodrich (1988) concluded that lentil should be as tall as possible at maturity for mechanical harvest to be efficient because more grain would be harvested.

1.5.2.4. Weeds

Weeds compete with crops for resources thus reduce crop yields. Lentil short stature and relatively slow early growth and late canopy closure make it sensitive to competition from weeds with faster growth potential (Yenish et al. 2009). A negative correlation was observed between weeds emergence and lentil grain yield (Kropff et al. 1992). Weeds can cause up to 60-100% grain losses (Wang, 2013). In conventional agriculture, weeds are controlled by herbicides but in organic agriculture where chemical inputs are prohibited, weeds can become an overwhelming issue if early mechanical weeding is not efficient. Lentil yields can also be reduced by late emerging weeds (Yenish et al. 2009).

Water stress, lodging, bruchid damage and weed competition contribute to the farmer's reluctance to grow lentils. It is important to develop alternative cropping managements that would help farmers by mitigating the agronomic constraints previously mentioned. Fortunately, an ancient cropping practice, particularly effective in organic farming, may be worth investigating.

1.6. Intercropping, an agronomic solution to lentil cropping constraints?

1.6.1. Definition and uses

Intercropping is the simultaneous growth of two or more species in the same field for a significant period but without necessarily sowing or harvesting at the same time (Willey 1979). It is an ancient, traditional, agricultural practice that is well established in some cropping systems of Latin America (e.g. beans intercropped with maize) or Africa where 98% of cowpeas

are intercropped but is near absent in Europe for human food production (Brooker et al. 2014). Several forms of intercrops exist worldwide such as mixed cropping (species mixed within rows without particular row arrangement) or alternate rows (species are grown in separate rows which are alternated).

1.6.2. General principles of intercropping

A plant within a cover interacts with other plants of the same species (intra-specific interactions) and plants of the other species (inter-specific interactions). The effectiveness of intercrops depends mainly on the management of competitions between plants for resource acquisition such as light, water or N, that are often limiting, especially in low inputs systems such as organic agriculture (Brooker et al. 2014; Bedoussac et al. 2014). In this context, complementary use of resources is a key factor for biomass and grain production of intercrops. This phenomenon occurs “when intercropped plants with complementary traits interact positively to increase productivity” (Brooker et al. 2014). The complementary use of N pools in legume-cereal intercrops thanks to the legume BNF has been largely reported in the scientific literature to be a key determinant of the better productivity of such intercrops (Peoples et al. 2009; Brooker et al. 2014; Bedoussac et al. 2014). Other forms of complementarities are known to be key factors of intercrop performance such as aerial and root architectures, complementarities for light and mineral resource acquisition (Brooker et al. 2014). Plants can also have a direct positive interaction with other plants in a process called “facilitation” which occurs for example when combining plants that increase the phytoavailability of resources such as water or nutrients (Brooker et al. 2014). Global performance of intercrops depends on many factors, such as the genotypes involved, plant densities, edaphic conditions, climate and interactions between all these factors (Brooker et al. 2014; Bedoussac et al. 2014).

1.6.3. Knowledge on lentil in intercrops

Several studies have shown promising results of intercropping lentil with cereals, especially when N availability is low, such as in organic farming (Yu et al. 2016). Intercropping has been shown to (1) increase total grain yield (Akter et al. 2004; Carr et al. 1995), (2) increase cereal grain protein concentration (Wang et al. 2013), (3) decrease weeds (Wang et al. 2013) and (4) improve higher gross margin (Akter et al. 2004). Altogether these results are promising but they are not sufficient to understand the functioning of intercrops and especially the interactions between species within the objective of producing grain and acquiring nitrogen. A better understanding of these intercrops would help to assess if this practice could be suitable for

organic lentil production in southwestern France and would more broadly contribute to the scientific knowledge on legume-cereal intercrops that can be a key to many agronomical locks worldwide.

1.6.4. Participation of the agricultural cooperative Qualisol in the thesis

This thesis is of the “Conventions Industrielles de Formation par la REcherche, (CIFRE)” type, i.e. a private organism participated in the funding of this work. In our case, it is the agricultural cooperative Qualisol which is based at Castelsarrasin, 70 kms northwest of Toulouse, in the department of Tarn-et-Garonne (82). It is a medium-sized cooperative that has a significant development in organic farming. The cooperative currently produces a large range of organic products, such as grain legumes (especially lentil) and cereal flour. The organic farming activity of the cooperative is flourishing, due to some extent to the sales of four lentil varieties, including the green “Anicia”, the cultivar of the renowned designation of origin “Lentille verte du Puy”. However, organic lentil production is dramatically constrained by the agronomic issues mentioned earlier. The cooperative Qualisol associated with the Institut National de la Recherche Agronomique (INRA) to investigate the potential of intercrops to mitigate these constraints and help increase lentil production. This thesis includes an applied research objective whose outcomes will be discussed in the final discussion of this report.

The selection of the companion crop for the lentil was made in concertation with the cooperative and INRA. The companion needed to be of economic interest for the cooperative and agronomically pertinent. We decided to use a cereal since intercrops of legume-cereal have been extensively studied in the scientific literature and have proved to be globally efficient. The synchronicity of the maturity period is considered as a decisive criterion for choosing companion crops in intercrop in the case of lentil because their pods become dehiscent and open upon ripening thus causing grain losses. In this context, spring wheat appears as a suitable candidate since its physiological maturity is concomitant with that of lentil and it has an economic interest for flour production for Qualisol. As for lentil, spring wheat is usually sown in early March in southwestern France to limit the risk of water stress during key phases of yield establishment i.e. booting and grain filling (Gate and Gouache 2010). The Risk of spring wheat lodging is low in organic farming since N is often limiting thus so the ability of wheat to act as a stake for the lentil should not be impeded.

1.7.Objectives of the thesis and main research questions

1.7.1. Main objectives, research questions and experimental strategy

This thesis has two main coupled objectives. The first one consists in investigating the performance of lentil spring-wheat intercrops to produce grain yield as compared to sole crops to decide if this agronomic practice is an interesting option to develop lentil production. The second is to understand the main underlying mechanisms that explain the performance to (1) design efficient cropping practices and (2) provide knowledge on lentil spring wheat intercrops that can be used for further research on this specific intercrop or more broadly on legume-cereals intercrops.

1.7.2. Main research questions

- Q1.** Do intercrops have an effect on grain yields as compared to sole crops?
- Q2.** Does the presence of wheat increase the mechanical harvestability of lentil?
- Q3.** Does the presence of wheat reduce the bruchid damage to lentil?
- Q4.** Is the grain protein concentration of crops affected by intercrops?
- Q5.** Do intercrops have higher gross margins compared to sole crops?
- Q6.** Can complementary use of resources explain the performance of intercrops?
- Q7.** How do intercrops affect species yield components and what are the main factors influencing competition intensities?

These questions have been examined in details in the following research chapters.

The experimental strategy to meet our objectives and research questions was to implement field experiments over two years at the experimental station of INRA Castanet-Tolosan. Several intercrop designs (additive and substitutive) were tested differing in cultivars and densities. Four lentil and two spring wheat cultivars, of economic interest, differing mainly on precocity (time from emergence to flowering), were tested. Variables measured to answer the research questions are detailed in the following chapters when needed.

1.8. Outline of the thesis

This thesis is composed of five chapters: the general introduction (Chapter I), three research chapters (Chapters II to IV), and a general discussion (Chapter V). The research chapters are in publication format with specific abstracts, objectives, introductions, material and methods, results, and discussions.

Chapter II: “Yield gap analysis extended to marketable grain reveals the profitability of organic lentil-spring wheat intercrops” investigates the effects of lodging and bruchid-beetles damage on lentil yields and crops gross margins, through an adaptation of the yield gap concept to include grain losses due to mechanical harvest and insufficient quality.

Chapter III: “Species niche complementarity for nitrogen improves lentil-wheat intercrop grain production in organic farming” focuses on the nitrogen use of intercrops to explain the agronomic performances of intercrops versus sole crops. This chapter also evaluates the effect of species densities within the intercrop on grain production.

Chapter IV: “Yield component analysis reveals that lentil-spring wheat intercrops grain yield is determined early during crop growth” examines the effects of interspecific competitions on yield components of lentil and spring wheat to better understand the dynamics of yield formation of species in intercrops.

Chapter V: “General discussion” sums up the novel information generated by our experimental work. The pros and cons of lentil spring-wheat intercrops are weighed within the framework of the thesis objectives. This chapter also includes perspectives of studies around lentil spring-wheat intercrops.

Chapter I. Yield gap analysis extended to marketable grain reveals the profitability of organic lentil-spring wheat intercrops

Viguier Loïc, Bedoussac Laurent, Journet Etienne-Pascal and Justes Eric

(2018). *Agronomy for Sustainable Development* 38–39. doi:10.1007/s13593-018-0515-5

Adapted from manuscript



Abstract

Lentil has been overlooked by organic farmers in Europe mainly because of low and unstable yields, notably due to lodging and bruchid beetles. Our study aimed to evaluate the efficiency of lentil-spring wheat intercrops to lower these reducing factors and increase yield and gross margin.

A two-year field experiment was carried out in southwestern France in 2015 and 2016 under organic farming rules. Four lentil and two wheat cultivars were grown as sole crops and intercrops. The “yield gap” concept was adapted to include grain losses due to mechanical harvest and insufficient quality.

Mean total intercrop grain yield before mechanical harvest was higher than mean sole crop (1.91 ± 0.47 vs. 1.57 ± 0.29 t ha⁻¹, respectively), with a lower mean yield of lentil in intercrop than in sole crop (1.06 ± 0.28 vs. 1.61 ± 0.54 t ha⁻¹). This led to a lower mean gross margin of intercrops than lentil sole crop (1772 ± 507 vs. 2371 ± 756 € ha⁻¹), before mechanical harvest. The percentage of bruchid-damaged grain did not differ significantly between intercrop and sole crop (41%). However, lentil lodging was lower in intercrop than in sole crop (15% vs. 40%), which strongly increased lentil mechanical harvest efficiency (75% vs. 50%). This led to a similar mechanically harvested yield of lentil in intercrop and sole crop (0.80 t ha⁻¹). Consequently, mean marketable gross margin of intercrops was higher than that of sole cropped lentil (949 ± 404 vs. 688 ± 393 € ha⁻¹), due to the addition of marketable wheat yield.

We thus demonstrated for the first time the interest of extending the yield gap concept to consider all grain losses that influence profitability, including those linked to mechanical harvest efficiency and insufficient grain quality. Furthermore, this is a first demonstration of the higher profitability of organic lentil-wheat intercrops compared to sole crops despite the additional costs associated with grain sorting.

Key-words: Lodging; Bruchid; Harvest efficiency; Gross margin

2.1. Introduction

Grain legumes are among the most common crop species in human diets worldwide (Erskine et al. 2011; Stefaniak and McPhee 2015). Lentil (*Lens culinaris* Med.) is a popular legume and an important source of protein in many countries in Asia and Africa, but not in Europe, where lentil consumption remains low despite recent increasing trends. Lentil is grown mainly in North America, southern and western Asia, North Africa (Erskine et al. 2016; Ghanem et al. 2015). Consequently, Europe imports a large percentage of the lentils its population consumes, which creates an opportunity for European producers, particularly organic producers, and for the development of agroecology and organic agriculture, which are forces driving the promotion of grain legumes (Erskine et al. 2016).

Lentil, a member of the *Fabaceae* family, can meet as a crop much (up to 80%) of its nitrogen (N) requirements through biological N₂ fixation, due to a symbiotic relationship between its roots and rhizobacteria (Reda 2015). This ability is particularly interesting in low N systems such as organic farming, in which N is often a limiting factor due to the prohibition of mineral fertilisers and the cost of organic ones. Introducing legumes into crop rotations is one way to increase sustainability, by increasing biodiversity, soil N fertility, and pest management at the cropping system level (Meynard et al. 2013; Voisin et al. 2013). Despite these potential advantages, grain legumes represented less than 2% of arable crop area in the European Union in 2014, of which lentil represented only 4.9% of the area dedicated to pulses (FAOSTATS, 2014). Farmers' reluctance to grow lentil can be explained in part by its low and unstable yields under European conditions. Intercropping is the simultaneous growth of two or more species in the same field for a significant period but without necessarily sowing or harvesting at the same time (Willey 1979). In the case of lentil-spring wheat intercrops, both species reach maturity at roughly the same time and are mechanically harvested together. Lentil grains are then separated from wheat grains using successive separating and cleaning tools such as vibratory, rotary, gravity and optical sorters. Like other legume-cereal intercrops, intercrops of lentil and wheat may be an interesting way to increase lentil production, as they have been shown to increase total yield (Akter et al. 2004; Carr et al. 1995) and gross margin (Akter et al. 2004).

Among challenges to lentil production, bruchid beetles (Coleoptera: Bruchidae) may decrease grain yields greatly, particularly in organic farming. Grain damaged by bruchids is not marketable and represents net yield and income losses for farmers that can exceed 50% (Laserna-Ruiz et al. 2012), especially in organic farming. In Europe, two established bruchid

species (*Bruchus lentis* and *B. signaticornis*) may cause great damage to lentil (Delobel 2005; Yus-Ramos et al. 2014). Adults lay eggs on the surface of developing pods, and larvae then penetrate the pod and feed on the growing lentil grain. These two species can damage lentil only in the field, as they are univoltine and do not lay eggs on stored grain (Yus-Ramos et al. 2014). No effective biocontrol method is currently available in the field for organic farmers, which hinders development of lentil in areas where bruchid damage is high. Meanwhile, to our knowledge, the potential of lentil-spring wheat intercrops to reduce the percentage of bruchid-damaged grains has never been studied, which is of particular interest for low-input systems without chemical control. Plant diversity can promote pest regulation through a phenomenon called “associational resistance” (Tahvanainen and Root 1972; Risch et al. 1983; Letourneau et al. 2011). Associational resistance is considered to occur because of two main ecological mechanisms (Root 1973; Andow 1991; Barbosa et al. 2009): (1) resource concentration, a bottom-up perspective predicting that pests are more likely to find and remain on host plants that are concentrated, such as in dense or nearly pure stands, and (2) natural enemies, a top-down perspective based on a positive correlation between plant species richness and natural enemy abundance. In lentil-spring wheat intercrops, we hypothesise that the spring wheat creates visual or olfactory confusion, decreasing the ability of bruchids to find pods for oviposition, thus reducing grain damage and financial loss (Kinane and Lyngkjaer 2003). This hypothesis does not exclude the potential control of pests by natural enemies.

Lodging is another common issue for lentil which results from a combination of genotypic and environmental factors (Ball et al. 2006). Lentil shoots collapse if unfavourable weather (e.g. rain and wind) occurs before harvest (Sidahmed and Jaber 2004). Impacts of lodging on grain yields may be limited in southern and western Asia, where most lentil is harvested by hand. However, in areas where lentil harvest is mechanical, such as North America or Europe, when lentil lodges, combine harvesters may fail to pick up plants leaning too much toward the ground, leading to large grain loss in the field, sometimes up to 100% (Carr et al. 1995). Even in countries where lentil is traditionally harvested by hand, mechanisation is gradually taking over because of increasing scarcity and costs of human labour (Erskine et al. 2016; Reda 2015), making them susceptible to reduced yields from lodging. Factors besides lodging influencing grain loss during mechanical harvest are numerous, such as weather conditions, field topography, ground roughness, management practices, crop maturity, type of combine harvester used and its settings and height of cut (Erskine and Goodrich 1988; Ibrahim et al. 1993). Erskine and Goodrich (1988) concluded that lentil should be as tall as possible at

maturity for mechanical harvest to be efficient because more grain would be harvested. The presence of wheat shoots in intercrops may act as stakes, keeping lentil shoots relatively upright and high (Carr et al. 1995; Erskine et al. 1991; Sidahmed and Jaber 2004); in this way, intercropping could decrease grain loss due to lodging by increasing mechanical harvest efficiency (Fig. 2).

Field pictures at harvest stage of sole cropped lentil, lentil-spring wheat intercrop and mechanical harvest of an intercrop



Fig. 2 (Left) sole cropped lentil and (middle) lentil-spring wheat intercrop at harvest, and (right) mechanical harvest of the intercrop. Orange marks on the ladders are spaced 10 cm apart. These pictures highlight that spring wheat in intercrops reduces lentil lodging at physiological maturity by maintaining plants relatively upright, allowing the combine harvester to pick up most shoots.

As increasing lentil production in Europe is desirable to lower importations and feed the increasing demand for organic products, it is important to provide information about agronomic issues and thus the economic feasibility of more sustainable agronomic solutions such as intercropping. Indeed, farmers are more likely to adopt new agricultural practices if they are economically promising or risk-limiting. In many studies, including several focusing on lodging and economic performance, lentil was hand-harvested. By not considering potential grain loss due to mechanical harvest, however, these studies may have overestimated yields, as mentioned by Wang et al. (2013), especially in areas where lentil would likely be mechanically harvested. Knowledge is lacking about effects of mechanical harvest with a combine harvester on sole cropped and intercropped lentil. In this study, our objective was to address three research questions: do lentil-spring wheat intercrops have higher total grain yield, mechanical harvest efficiency, and/or profitability than sole cropped lentil in organic farming? We thus developed an original approach to analyse the issues of lodging and bruchids in lentil, by adapting the “yield gap” concept developed by Evans (1994) and revised by Van Ittersum et al. (2013). Yield gap analysis identifies and quantifies “limiting” and “reducing” factors of a crop (Van Ittersum et al. 2013). In our adaptation, we added two downstream stages to estimate all grain losses down to the “marketable” yield.

2.2. Materials and methods

2.2.1. Improving the yield gap concept

The yield gap concept was adapted by adding two downstream stages to the established yield gap sequence to estimate all grain losses from “attainable” yield down to “marketable” yield, which is composed of only grain that can be conditioned and sold for human consumption (Fig. 3). Attainable yield is that obtained in the presence of limiting factors (e.g. water, N). “Actual” yield is the yield after the occurrence of biotic reducing factors and is estimated by hand-harvest, assuming that all grain produced in the field is collected. In this study, actual yield is composed of three distinct fractions: (1) sound, marketable grain, (2) bruchid-damaged, non-marketable grain and (3) “small grain”, non-marketable for human consumption. “Mechanically harvested” yield, which corresponds to actual yield minus grain loss in the field during mechanical harvest, is also composed of sound grain, bruchid-damaged grain, and small grain. Finally, marketable yield corresponds to mechanically harvested yield minus grain discarded after the sorting process because it falls below quality and sanitary standards (i.e. small grain and bruchid-damaged grain in our study). Note that we hereafter call “sorting process” the combination of both the separation of grains from the two intercropped species (whenever relevant) and the cleaning process where all contaminants, debris and below-standard grain fractions are discarded. Separation and cleaning occur at the same time on the grain sorting chain.

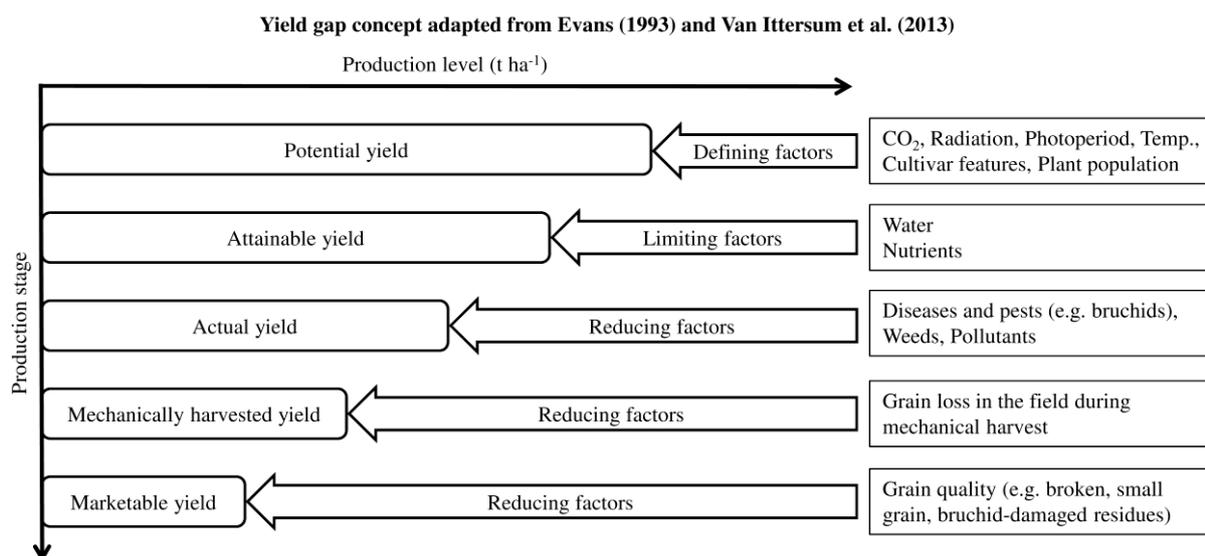


Fig. 3 Extension of the yield gap concept to the marketable yield. Agronomic production is influenced by growth-defining, limiting and reducing factors. Attainable yield is that obtained in the presence of limiting factors. Actual yield is that obtained after the occurrence of biotic reducing factors and when a crop is hand-harvested. Two subsequent production stages were added: (1) mechanically harvested yield, which equals actual yield minus a reducing factor due to loss in the field during mechanical harvest, and (2) marketable yield, which is composed of only human-edible grain and corresponds to mechanically harvested yield minus a reducing factor due to discarding grain that falls below quality and sanitary standards.

2.2 Site, soil and climate

A two-year field experiment was carried out at the Institut National de la Recherche Agronomique station in Auzeville – southwestern France, 43°31'N, 1°30'E – in 2015 and 2016. In 2015, soil was sandy clay loam (25% clay, 23% silt and 52% sand) with total soil water content at sowing of 294 mm (0-120 cm soil depth). In 2016, soil was loam (30% clay, 30% silt and 40% sand) with total soil water content at sowing of 286 mm (0-120 cm). Soil mineral N at sowing was low, with 31 and 20 kg N ha⁻¹ in 2015 and 2016, respectively (0-120 cm). The sum of daily mean temperatures over the growing period was above the 20-year mean of the experimental site (3060 vs. 2901 °C day, respectively, on a 0°C basis) in 2015, but lower (2766 °C day) than it in 2016. Moreover, mean daily maximum temperature during a key lentil developmental period – from flowering to maturity – was higher in 2015 than in 2016 (29°C vs. 26°C, respectively). Total rainfall during the growing period was similar in both years (305 and 286 mm in 2015 and 2016, respectively) and similar to the 20-year mean (282 mm). Rainfall in 2015 had heterogeneous distribution, however, with 57% of that during the growing period concentrated in only two storm events (early May and mid-June), without any rain in between. These conditions may have led to water stress for plants from mid-May to mid-July.

Conversely, rainfall in 2016 had homogenous distribution throughout the growing period, and we assumed that no water stress occurred.

2.2.2. Experimental design

The experiment was a randomised block design with three replicates. Four lentil cultivars (cv.) – Anicia, Beluga, Flora and Rosana, yielding green, black, yellow and red grains, respectively – and two spring wheat (*Triticum aestivum*) cultivars – Valbona and Togano – were each grown as (1) sole crops with an objective of 300 and 450 plants m⁻² for lentil and wheat, respectively, and as (2) two-species intercrops – in 8 cultivar pairs – in a partial additive design with a plant density ratio of 100% of sole cropped lentil:17% of sole cropped spring wheat. Spring wheat's low plant density in intercrop was chosen to limit its competition with lentil. Lentil was planted at 100% density to maximise its yield in intercrop and thus the intercrop's profit, given the much higher price of lentil compared to that of wheat. Crops were sown on 12 March 2015 and 23 March 2016. Each plot consisted of 10 rows, 10 m long in 2015 and 8 m long in 2016, spaced 16.5 cm apart. In intercrop plots, the two species were homogeneously mixed within each row to maximise the ability of wheat shoots to act as stakes. Mean plant density after emergence among all treatments reached 95% in 2015 and 101% in 2016 of the plant density objective. The experiment was conducted under organic farming rules; thus, neither synthetic pesticides nor chemical fertilisers were applied. Besides bruchids on lentil, no other significant yield-reducing biotic factors such as diseases or weeds were observed on any of the sole crops or intercrops.

2.2.3. Measurements, calculations and statistics

2.2.3.1. Actual yield measured by hand-harvest

To avoid edge effects, an area of 1.98 m² (2 m long, 0.99 m wide) of the six inner rows of each plot was hand-harvested at lentil maturity, around mid-July in both years. Spring wheat always reached maturity before lentil, but no wheat grain was lost despite the delay in harvest, due to its indehiscence. Lentil shoots were carefully hand-harvested to prevent pod opening and grain loss and attached root fragments were discarded. Spring wheat was cut at ground level. Crops were threshed separately using a research-designed thresher (brand Roland Chateau du Loir, France) ensuring little grain damage or loss. Grain was then processed through an air separator to separate heavier sound grain from lighter grain (i.e. small grain and, for lentil, bruchid-

damaged grain). Grain fractions were subsequently oven-dried for 48 hours at 80°C for dry weight determination.

2.2.3.2. Mechanical harvest efficiency measurement

To estimate lentil grain loss in the field during mechanical harvest using a combine harvester, an experiment was performed in 2016 with lentil cv. Anicia, both in sole crop and in intercrop with spring wheat cv. Valbona. It consisted of two identical lines of plots in which treatments were randomly placed and replicated three times, contiguous to those used previously to estimate the actual yield by hand-harvest. In one line, crops were hand-harvested following the same protocol as previously described, and in the other line, crops were mechanically harvested with a research-designed combine harvester as a prototype of classic combine harvesters (Fig. 3). The combine harvester cutter-bar was placed as low as mechanically possible – i.e. 5 cm above the ground – to simulate farming practice conditions. Machine settings – sieve size and fan speed – were first calibrated in additional dedicated lentil plots to maximise mechanical harvest efficiency. The area mechanically harvested by the combine harvester was measured for each plot (mean = 11 m²). Grain dry weight was determined as for hand-harvested plots, except that grain from the combine harvester was already threshed and only had to be air-cleaned. Finally, mechanical harvest efficiency was calculated as a function of the fraction of sound grain.

2.2.3.3. Lentil height, stem length, lodging and lowest pod height at harvest

Lentil plant height (cm) at harvest was measured one day before harvest in each hand-harvested plot using a ruler placed vertically at six randomly chosen places in the six inner rows. Lentil stem length (cm) was defined as the distance from the collar to the top of slightly stretched-until-straight shoots. Each lentil plant sampled for estimating bruchid-damaged grain in 2016 was measured, and the lodging (%) was estimated as follows:

$$\text{Lodging} = \left(\frac{\text{Lentil stem length} - \text{Lentil height at harvest}}{\text{Lentil stem length}} \right) \times 100$$

Height of the lowest lentil pod was measured in 2016 for lentil cv. Anicia in sole crop and in intercrop with spring wheat cv. Valbona in each plot of the hand-harvested line of plots used to estimate mechanical harvest efficiency, on one plant – randomly chosen – every 10 cm over a 50 cm-long row segment. This operation was repeated three times per plot.

2.2.3.4. Estimation of bruchid-damaged grain rate, total grain loss due to bruchids and attainable yield

Evaluating grain loss due to bruchids by using insecticide-sprayed plots as a control was not considered an option for our experiment, since the plots were too small to prevent dispersal of bruchids from non-sprayed plots to sprayed plots, even with repeated spraying. Therefore, we estimated grain loss due to bruchids by measuring the percentage of damaged grains in each plot and the mean one-grain mass of bruchid-damaged grains. Twenty plants – five times four consecutive plants in the six inner rows – were collected from each hand-harvested plot. Plants from each plot were manually threshed, and the grain was immersed in a basin of water. Sound grain sinks to the bottom, while bruchid-damaged grain floats to the surface. The water was stirred to separate the grains completely. A sieve was used to collect floating grain and then submerged grain. Each grain that had floated was then pressed with a finger to confirm that it was bruchid-damaged, as bruchid-damaged grain cracks when pressed. Bruchid-damaged and sound grains were then dried and counted separately using a grain counter. This original method provided quick and accurate estimates of the bruchid damage rate for a large sample of grain and also the ability to detect bruchid-damaged grain from which adult insects had not yet emerged, something which cannot be done visually. Sound and bruchid-damaged grain from 16 plots in 2015 were oven-dried for 48 hours at 80°C to determine the one-grain weight for each lentil cultivar.

The total grain mass lost due to bruchids (t ha^{-1}) equalled the sum of (1) the mass of bruchid-damaged grain residues (measured) and (2) the mass of grain consumed by bruchids (unknown). It was estimated for each plot as follows:

$$\begin{aligned} & \text{Total grain mass lost due to bruchids} \\ &= \text{Number of bruchid-damaged grains} \times \text{Mass of one sound grain} \\ &= \frac{\text{Mass of bruchid-damaged grain residues}}{\text{Mass of one bruchid-damaged grain}} \times \text{Mass of one sound grain} \end{aligned}$$

The mass of grain consumed by bruchids (t ha^{-1}) for each plot was calculated as follows:

$$\begin{aligned} & \text{Mass of grain consumed by bruchids} \\ &= \text{Total grain mass lost due to bruchids} - \text{Mass of bruchid-damaged grain residues} \\ &= \text{Mass of bruchid-damaged grain residues} \times \left(\frac{\text{Mass of one sound grain}}{\text{Mass of one bruchid-damaged grain}} - 1 \right) \end{aligned}$$

Finally, the attainable yield (t ha^{-1}) for each plot was calculated as the sum of the actual yield and the mass of grain consumed by bruchids after cleaning process. In support of these equations, we assumed that bruchid larvae damaged developing lentil grains late enough during the crop reproductive phase so that lentil plants could not compensate for the damage and that lentil yield components (e.g. number of grains, mass of one sound grain) remained similar to those achieved in the absence of bruchids.

2.2.3.5. Economic parameters

Actual and marketable gross margins (€ ha^{-1}) were calculated for both lentil and spring wheat in both sole crop and intercrop, considering the actual and marketable yields (t ha^{-1}) respectively, as follows:

Gross margin

$$= \text{grain yield} \times \text{selling price} - \text{sowing seed quantity} \times \text{seed cost} - \text{grain yield} \times \text{sorting cost}$$

For intercrops, we then summed the gross margins of both lentil and spring wheat. In calculations, we considered only the selling price, seed cost and sorting cost – costs that differed between crops – which corresponds formally to a partial gross margin. For simplification, however, we hereafter use "gross margin". Note that, for the actual yield – composed of sound and damaged grains – our calculation of gross margin assumes that damaged grains are not removed and therefore sold at the same price as sound grains. We deliberately included these damaged grains in actual yield calculation to further demonstrate that extending the yield gap concept to consider all grain losses that influence profitability, including those due to mechanical harvest and damaged grain disposal, is essential to reveal the real profitability of lentil spring wheat intercrops.

Seed costs and grain selling prices were provided by the agricultural cooperative Qualisol, located in southwestern France, which commercialises lentil produced by farmers in both sole crop and intercrop. Seed costs were 3150 € t^{-1} for lentil cv. Anicia, Flora and Rosana; 6000 € t^{-1} for lentil cv. Beluga and 1030 € t^{-1} for each spring wheat cultivar. Selling prices were 1792 € t^{-1} for lentil cv. Anicia, Flora and Rosana; 2800 € t^{-1} for lentil cv. Beluga; 448 € t^{-1} for sole cropped spring wheat and 504 € t^{-1} for intercropped spring wheat because its protein content exceeded 14%. Note that Beluga has higher market demand but lower potential yield than those of other lentil cultivars, which justifies its relatively higher seed cost and selling price. To estimate grain sorting costs, we used those also furnished by the agricultural cooperative

Qualisol: 11, 11, 45 and 67 € t⁻¹ per pass of, respectively, rotary cleaner, vibratory separator, gravity separator and optical sorter. Note that even when sole cropped, lentil needs to be thoroughly sorted (2 passes of each tool for a total of 268 € t⁻¹) to remove all stones, dust, and broken and bruchid-damaged grain from marketable grain. Intercrop grain mixtures are sorted similarly to sole cropped lentil but with 4 passes of optical sorter (total of 402 € t⁻¹). As a comparison, the sole cropped wheat grains only need 1 pass of each cleaner/separator and no optical sorting (total of 67 € t⁻¹).

2.2.3.6. Statistical analysis

Shapiro-Wilk and Levene's tests were used to test the normality of the data and the homoscedasticity of its variance, respectively. Pairwise *t*-tests were used to compare treatments for all dependent variables (e.g. grain yield, N accumulated) using the "t.test" function of R software via Rstudio (version 1.0.136). If necessary, data were square log-transformed to obtain a normal distribution. Unequal variance was accounted for in the *t*-test if Levene's test indicated heteroscedasticity. When possible, one-tailed *t*-tests were performed. All results are presented as mean ± 1 standard deviation.

2.3. Results and discussion

2.3.1. Effect of intercrops on actual yield

Considering all cultivars together, the mean actual yield of intercrops (lentil + spring wheat) was significantly higher than that of sole cropped lentil (Fig. 4a) in both 2015 (1.57 and 1.29 t ha⁻¹, respectively; *P*<0.01) and 2016 (2.26 vs. 1.93 t ha⁻¹, respectively; *P*<0.01). Mean actual yield of sole cropped spring wheat (1.46 and 1.60 t ha⁻¹ in 2015 and 2016, respectively) was less than or equal to that of intercrops in almost all treatments, but the difference was significant only in 2016 (*P*=0.26 and *P*<0.001 for 2015 and 2016, respectively). There was no significant effect of cultivar or year on actual yield of spring wheat, allowing actual yields of the two wheat cultivars to be averaged together. Although actual yields of lentil included bruchid-damaged grain and small grain, the trends observed were the same when only the sound grain fraction of actual yield was considered (data not shown). Note that the amount of small grain turned out to be negligible.

These results indicate a grain yield advantage in intercrop vs. in sole crop under a wide range of conditions: two years with contrasting climates, four lentil varieties and two wheat varieties. This increase led to a land equivalent ratio (LER) – the relative land area of sole crops required

to produce the same yield achieved in intercrop, and with the same species proportion in total grain (Willey and Osiru 1972) – ranging from 1.02-1.54 (mean = 1.24 ± 0.14) based on the actual yield. The LER illustrates the ability of lentil-spring wheat intercrops to increase total yields in low-input systems and organic farming, as reported in other studies (Carr et al. 1995) and for other legume-cereal intercrops (e.g. Bedoussac et al. 2015; Fletcher et al. 2016). The intercrop's better performance can be explained by complementary use of N niches by lentil and spring wheat. As cereal forces legume to meet more of its N requirements by fixing N_2 (e.g. Bedoussac et al. 2015), the lentil does not totally compete with spring wheat for soil mineral N when intercropped (e.g. Naudin et al. 2009). Furthermore, our results suggest that the lower the yield of sole cropped lentil, the higher the yield advantage in intercrop (Fig. 4a), indicating that this species mixture could also be a way to ensure a minimum grain yield for organic farmers among years, especially when lentil yields are low, for example due to dry spring conditions. Moreover, when lentil yields were high, intercrops produced more than sole cropped spring wheat, probably because N was limiting in both experimental years (Tosti et al. 2016). Our results also agree with those of Bedoussac and Justes (2010), who observed that total grain yields of cereal-wheat intercrops were higher than those of sole cropped wheat when N availability remained low, such as in stockless organic farming.

Mean actual yield of lentil was significantly lower in intercrop than in sole crop (Fig. 4b) in both 2015 (0.93 vs. 1.29 t ha⁻¹, respectively; $P < 0.01$) and 2016 (1.20 vs. 1.93 t ha⁻¹, respectively; $P < 0.001$). This highlights that spring wheat added at a low density (17% of sole crop density) was still dense enough to decrease the associated lentil yield, illustrating strong interspecific competition of spring wheat with lentil. Similar trends were observed in several previous studies of lentil-wheat intercrops (Akter et al. 2004; Carr et al. 1995; Wang et al. 2013). Actual yield of lentil in sole crop and intercrop was significantly higher in 2016 than in 2015 ($P < 0.001$ and $P < 0.01$, respectively; Fig. 4b). Actual yield of intercropped lentil tended to be higher when that of sole cropped lentil was high, i.e. when conditions were favourable for lentil growth. These results can be explained in part by favourable temperature and rainfall conditions around flowering and early pod filling stages in 2016, greatly increasing the number of pods per plant (data not shown) and thus the yield, unlike in 2015, which had a dry spring.

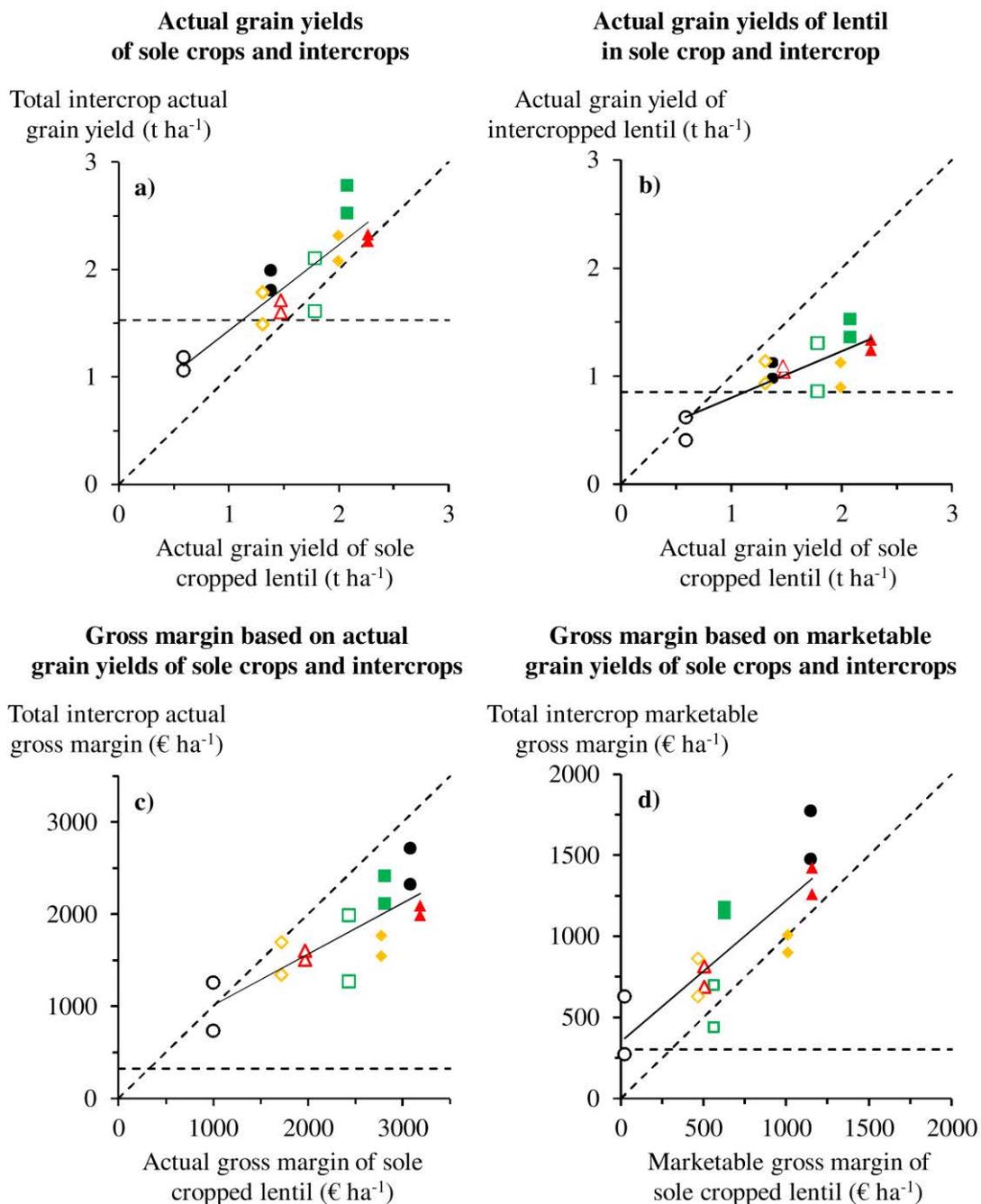


Fig. 4 Yields and gross margins of lentil-spring wheat intercrops compared to those of sole cropped lentil. a) Actual grain yield (t ha⁻¹) of the intercrop (lentil + spring wheat) vs. that of sole cropped lentil ($y=0.80x+0.63$; $R^2=0.78^{***}$); b) Actual grain yield of intercropped lentil (t ha⁻¹) vs. that of sole cropped lentil ($y=0.43x+0.37$; $R^2=0.63^{***}$); c) Actual gross margin (€ ha⁻¹) of the intercrop (lentil + spring wheat) vs. that of sole cropped lentil ($y=0.54x+458$; $R^2=0.64^{***}$); d) Marketable gross margin (€ ha⁻¹) of the intercrop (lentil + spring wheat) vs. that of sole cropped lentil ($y=0.87x+351$; $R^2=0.67^{***}$). *** indicates P-values<0.001. Symbol color and shape indicates the lentil cultivar (green square = Anicia, black and circle = Beluga, orange diamond = Flora, red triangle = Rosana). Symbol filling indicates the experimental year (open = 2015, closed = 2016). N=16. Dashed horizontal lines indicate the mean yield or gross margin of spring wheat in sole crop (both cultivars and years combined) for Figs. 4a, 4c, 4d or in intercrop (Fig. 4b).

2.3.2. Effect of intercropping on gross margin from actual yield

Mean actual gross margin of intercrops was significantly lower than that of sole cropped lentil but higher than that of sole cropped wheat (Fig. 4c) in 2015 (1427, 1778 and 304 € ha⁻¹, respectively; $P < 0.05$) and 2016 (2117, 2965 and 346 € ha⁻¹, respectively; $P < 0.001$). Despite the higher total yield of intercrops, the decrease in lentil yield in intercrop compared to that in sole crop was not economically offset by the actual yield of spring wheat in intercrop, given a selling price of lentil ca. four times that of spring wheat. Therefore, as lentil contributes more to intercrop gross margin, one should favour lentil yield to maximise gross margin of the actual yield of intercrops. Our results show that when hand-harvested – which corresponds to the actual yield – intercrop is less profitable than lentil sole crop. Finally, actual gross margins ranged widely over the two years of experiments, from 734-2715 € ha⁻¹ for intercrops and 1002-3188 € ha⁻¹ for sole cropped lentil. Thus, intercrops with lentil can achieve high, albeit lower, actual gross margins, even with a strong decrease in the actual yield of lentil. Akter et al. (2004) observed an economic advantage in the actual yield of lentil-wheat intercrops for management strategies including irrigation, fertilisation and chemical control of biotic stresses. However, since lentil is intended to human food and harvested with combine harvesters one should include the potential grain losses due to non-edible seeds (e.g. bruchid-damaged grains) and losses on field due to mechanical harvest to reveal marketable yield and marketable gross margin that reflect more accurately the reality of farmers.

2.3.3. Effects of intercrops on bruchid damage, lodging and mechanical harvest efficiency

2.3.3.1. Effect of intercrops on bruchid damage

Among all cultivars and years, mean percentage of bruchid-damaged grain was not significantly different for lentil in intercrop and sole crop (40±15% vs. 42±14%, respectively). Both treatments had a high mean percentage of bruchid-damaged grain in 2015 (49%) and a lower one in 2016 (33%). No difference in the bruchid damage was observed among cultivars except for Anicia, which was more sensitive (mean = 63% and 52% in 2015 and 2016, respectively, for sole crop and intercrop combined). Leroi et al. (1990) observed no significant difference in bruchid damage to cowpea intercropped with maize and that in sole crop. In contrast, Karel et al. (1982) and Olubayo and Port (1997) observed a significant decrease in bruchid infestation rate in cowpea-maize intercrops. These studies were carried out in East Africa, which has different bruchid species than those established in Europe. This result emphasises, however,

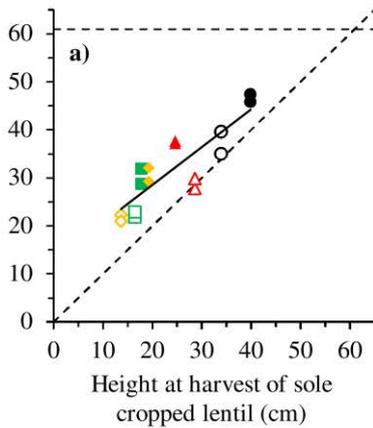
that increased plant diversity in the field can decrease bruchid infestation rate. Under our conditions, we estimated that mean yield loss due to bruchids was 0.69 and 0.93 t ha⁻¹ in 2015 and 0.52 and 0.93 t ha⁻¹ in 2016 for lentil in intercrop and sole crop, respectively. The presence of *B. lentis* and *B. signaticornis* has been confirmed in southwestern France (Yus-Ramos et al. 2014), but to our knowledge this is the first report of major damage by bruchids in this area in a scientific publication. Currently, the abundance of bruchids in southwestern France, coupled with the lack of effective agronomic or biological methods to control them, seriously hinders development of lentil in organic agriculture there. Unfortunately, our experiment cannot help to identify factors influencing bruchid damage, as it was not designed to do so, and no clear trend in damage was observed. Moreover, bruchid ecology is not well known, but we can hypothesise that bruchid infestations are influenced by temperature or degree-days during the growing season, as well as by crop rotations, landscape pattern and biodiversity.

2.3.3.2. Effect of intercrops on lentil height at harvest, stem length and lowest pod height

Mean lentil height at harvest (Fig. 5a) was higher in intercrop than in sole crop, non-significantly in 2015 (28 vs. 23 cm, respectively; $P=0.23$) but significantly in 2016 (36 vs. 25 cm, respectively; $P<0.01$). Akter et al. (2004) observed a similar increase in lentil height in intercrop. Mean lentil height at harvest in intercrop was lower in 2015 than in 2016 ($P<0.05$), while no difference was observed in sole crop ($P=0.38$). In 2016, mean lentil stem length (Fig. 5b) was similar between intercrop and sole crop (42 cm; $P=0.75$). Thus, the mean lodging was 15% in intercrop and 40% in sole crop (Fig. 5c). These results suggest a strong decrease in lentil lodging due to having spring wheat in the intercrop. Furthermore, mean height of the lowest pod was also significantly higher in intercrop than in sole crop (22 vs. 12 cm, respectively; $P<0.001$; data not shown). Thus, sowing wheat at 17% of its sole cropped density in intercrop was sufficient to significantly increase lentil height and lowest pod height at harvest. Moreover, the lower the height of sole cropped lentil at harvest, the larger its difference with the height of intercropped lentil (Fig. 5a). Carr et al. (1995) observed an increase of 3.5 cm in the height of the lowest lentil pod (albeit smaller than our result) in intercrop compared to that in sole crop. Thus, intercrops could be a way to significantly decrease lentil lodging, thus increasing pod height and creating conditions in which combine harvesters are more likely to gather more of the actual yield of lentil.

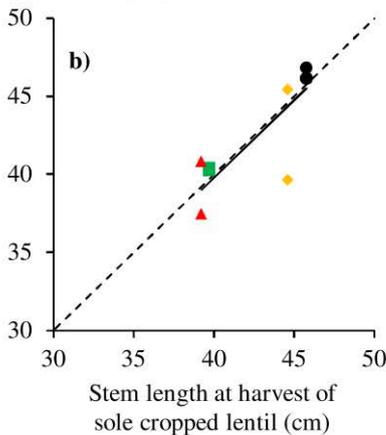
Lentil height at harvest in intercrop as a function of that in sole crop

Height at harvest of intercropped lentil (cm)



Lentil stem length at harvest in intercrop as a function of that in sole crop

Stem length at harvest of intercropped lentil (cm)



Lentil lodging rate in intercrop as a function of that in sole crop

Lodging at harvest of intercropped lentil (%)

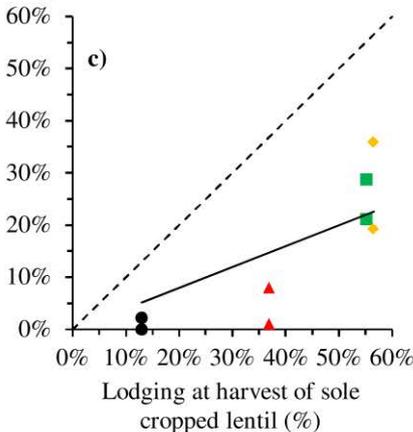


Fig. 5 Height, stem length and lodging of lentil intercropped with spring wheat compared to those of sole cropped lentil. a) Height at harvest (cm) of intercropped vs. sole cropped lentil ($y=0.79x+12.7$; $R^2=0.75^{***}$); b) Stem length at harvest (cm) of intercropped vs. sole cropped lentil ($y=0.99x$; $R^2=0.61^{**}$); c) Lodging at harvest (%) of intercropped vs. sole cropped lentil ($y=0.40x$; $R^2=0.61^{**}$). * represent P-values, ** when $P<0.01$ and *** when $P<0.001$. Intercropped lentil height at harvest exceeded that of sole cropped lentil in almost all treatments (mean = 32 vs. 24 cm, respectively), while lentil stem length was similar (mean = 42 cm). Lentil intercropped with spring wheat had a lower lodging than sole cropped lentil (mean = 15% vs. 40%, respectively). Symbol color and shape indicates the lentil cultivar (green square = Anicia, black circle = Beluga, orange diamond = Flora, red triangle = Rosana). Symbol filling indicates the experimental year (open = 2015, closed = 2016). $N=16$. The dashed horizontal line indicates the mean yield of intercropped spring wheat (both cultivars and years combined).

2.3.3.3. Effect of intercrops on mechanical harvest efficiency

Our mechanical vs. hand-harvest experiment performed in 2016 (Fig. 6) confirmed that the mean hand-harvested yield of sound lentil grain was lower in intercrop than in sole crop (1.01 ± 0.19 vs. 1.29 ± 0.13 t ha⁻¹, respectively; $P < 0.05$). In contrast, the mean yield of mechanically harvested lentil was similar in intercrop and sole crop (0.75 ± 0.11 vs. 0.64 ± 0.06 t ha⁻¹, respectively; $P = 0.81$). Consequently, mechanical harvest efficiency was clearly higher for lentil in intercrop than in sole crop (75% vs. 50%, respectively, $P < 0.05$). The greater mechanical harvest efficiency in intercrop can be attributed mainly to the higher mean pod height in intercrop, confirming the importance of maintaining pod height as high as possible. The slight increase in the lowest pod height observed by Carr et al. (1995) decreased grain loss of lentil in intercrop by only 3% compared to that in sole crop. They provided no data on lentil lodging, however, making comparison with our experiment impossible. Breeding lentil cultivars for high mechanical harvest efficiency appears to be a viable long-term strategy for issues related to mechanical lentil harvest. Moreover, it would be interesting to determine the minimum relative density of spring wheat needed to increase mechanical harvest efficiency of lentil in intercrop and simultaneously decrease its strong interspecific competition with lentil. We hypothesise that densities below 17% of its sole crop density can reach these objectives, notably due to the ability of spring wheat to compensate for low density by growing more shoots. However, reducing wheat density at sowing could increase at least two risks: (1) that farmers would fail to obtain good spatial distribution of wheat seeds, even using a pneumatic precision drill, and (2) that unfavourable climatic conditions would decrease wheat density even further by decreasing its emergence rate.

Mechanical harvest efficiency of lentil in sole crop and intercrop

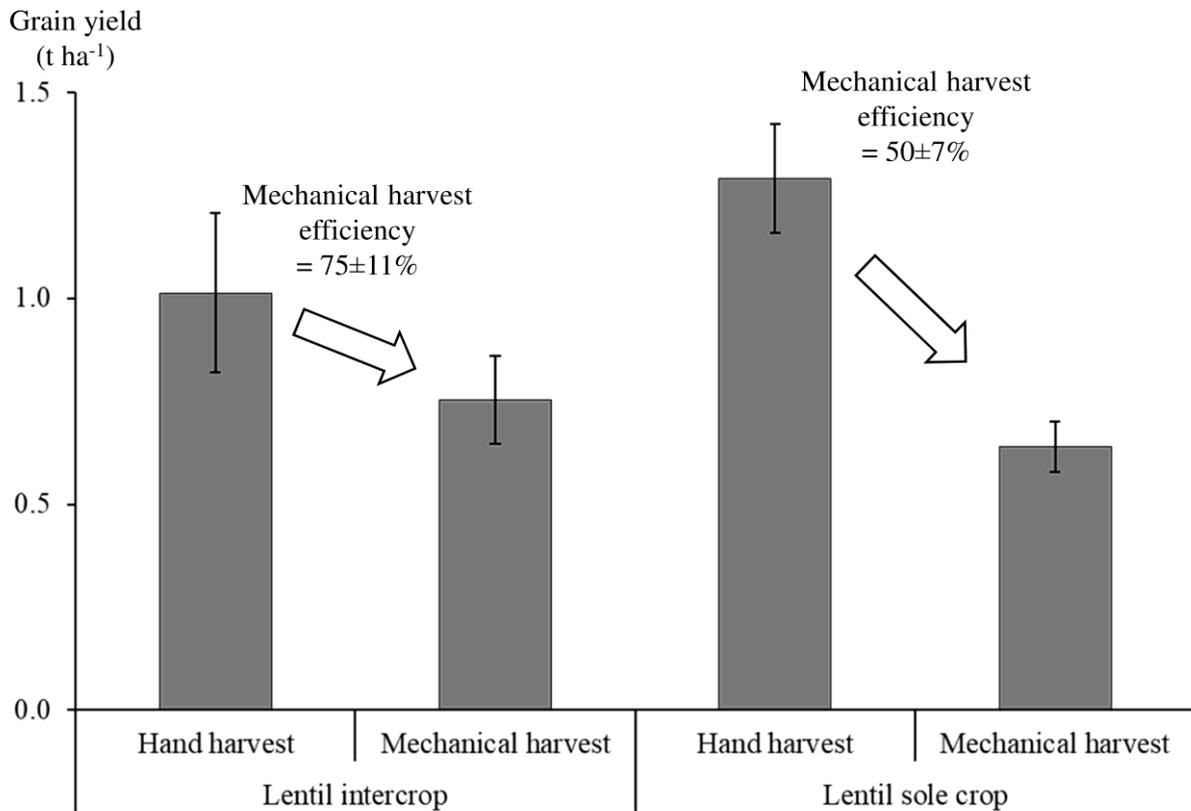


Fig. 6 Mean harvested yields of hand-harvested and mechanically harvested lentil in intercrop and sole crop, used to calculate mechanical harvest efficiency. The field experiment was performed in 2016 with lentil cv. Anicia and spring wheat cv. Valbona, considering only sound grain (N=3 for each). Mechanical harvest efficiency was higher for lentil in intercrop than in sole crop, leading to similar yields of sound grain. Error bars represent 1 standard deviation.

2.3.4. Effect of intercrop gross margin from marketable yield

We applied mechanical harvest efficiency to the actual yields to estimate mechanically harvested yields. We first assumed that mechanical harvest efficiency was the same for all lentil cultivars in both years, which seemed acceptable based on our observations of lentil height at harvest and stem length. These observations suggested that, even though mechanical harvest efficiency can vary among cultivars and years, the relative difference in mechanical harvest efficiency between lentil in intercrop and sole crop remains large. We then assumed that loss of spring wheat grain during mechanical harvest was negligible, as confirmed by our field observations after harvest, and did not significantly affect marketable gross margins. Next, we assumed that mechanical harvest efficiency was the same for all grain fractions of actual yield (i.e. marketable, bruchid-damaged and small). Finally, the marketable yield was used to calculate marketable gross margin to compare intercrop vs. sole crop profitability.

Mean marketable gross margin (Fig. 4d) was significantly higher for intercrops (lentil + spring wheat) than sole cropped lentil or sole cropped wheat in both 2015 (629, 390 and 283 € ha⁻¹, respectively; $P<0.05$) and 2016 (1269, 987 and 325 € ha⁻¹, respectively; $P<0.05$). Furthermore, marketable gross margins, like actual gross margins, ranged widely over the two years (273-1773 and 23-1158 € ha⁻¹ for intercrops and sole crops, respectively). The lowest marketable gross margin of intercrops was higher than that of sole cropped lentil ($P<0.05$), meaning that intercrops can act as “harvest insurance” for farmers, especially when sole crop yields are low. On the other hand, the highest marketable gross margin of intercrops was also higher than that of sole cropped lentil ($P<0.001$). Consequently, when lentil yield is high in sole crop, intercropping lentil may still be a way to increase gross margins. Intercropped lentil was thus found to be more profitable than sole cropped lentil in our experiments, under both favourable and unfavourable climatic conditions in organic farming. The decrease in lentil lodging due to support by wheat is an example of the “within-season benefit” concept developed by Fletcher et al. (2016) and helped to assess agronomic and economic performances of intercrops.

2.3.5. Yield gap analysis of all cultivars and years combined

Finally, we used our adaptation of the yield gap concept to detail lentil grain losses along the agronomic production stages in sole crop and in intercrop, for all cultivars and years combined. Mean attainable yield was 1.41 and 2.14 t ha⁻¹ for lentil in intercrop and sole crop, respectively (Fig. 7). Mean attainable yield of sole cropped lentil was high and even higher with cv. Anicia in 2016 (3.11 t ha⁻¹). This yield is consistent with that (3.0 t ha⁻¹) observed by Wang et al. (2013) in an experiment conducted with cv. Anicia in organic farming in Germany without water stress. This strengthens our assumption that our growing conditions were favourable (i.e. no water stress) for lentil in 2016.

Although bruchids consumed ca. 25% of the attainable yield of lentil in both intercrop and sole crop, we observed a mean actual yield in both intercrop and sole crop that was relatively higher than the mean worldwide lentil yield (ca. 1.0 t ha⁻¹, Erskine et al. 2011). Subsequently, 25% and 50% of the actual yield was lost during the mechanical harvest of lentil in intercrop and sole crop, respectively. Finally, a large mass of bruchid-damaged grain residues, representing 25% of the mechanically harvested yield of lentil in both intercrop and sole crop, had to be removed from the mechanically harvested yield to obtain the marketable yield (Fig. 7). Note that additional downstream stages can be added if higher grain quality is required by agro-food industries.

Ultimately, the marketable yield of lentil in intercrop was only 42% of its attainable yield but was higher than that in sole crop, which was only 28% of its attainable yield. Intercropped lentil approaches attainable yield more closely than sole cropped lentil (and with less risk), but both systems currently lay far below optimum performances. The yield gap analysis (Fig. 7) illustrates that grain loss at mechanical harvest was an important issue for lentil but clearly highlights that bruchids were the major reducing factor in our experiments, as is the case for organically farmed lentil in southwestern France.

Yield gap concept developed for lentil in sole crop and intercrop with spring wheat

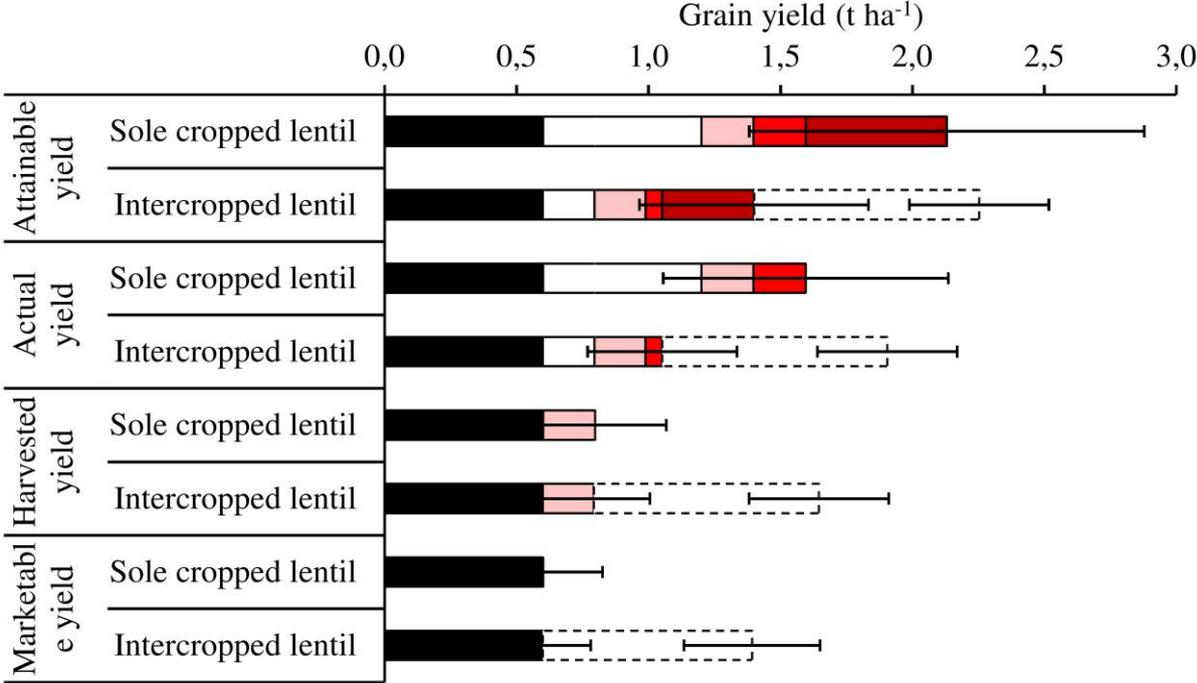


Fig. 7 Yield gap analysis for lentil intercropped with spring wheat and sole cropped lentil. For all cultivars and years combined, total grain loss from attainable to marketable yield was 58% for intercropped lentil and 72% for sole cropped lentil, leading to similar marketable yields. Bars indicate the mean mass of grain yield or loss for a given production stage: dark red bars for grain eaten by bruchids; red bars for bruchid-damaged grain residues eliminated from actual yield during mechanical harvest; light red bars for bruchid-damaged grain residues discarded by the final grain cleaning process respectively; white bars for sound grain lost during harvest; dashed-outline bars for wheat grain and black bars for the marketable yield of lentil. The mass of small grain lost during mechanical harvest and cleaning stages is not represented because of its insignificant weight compared to those of the other grain fractions. N=16 for intercropped lentil and N=8 for sole cropped lentil. Error bars represent 1 standard deviation.

2.4. Conclusion

This study illustrates the ability of lentil-spring wheat intercrops to yield more total grain than sole cropped lentil in low-input organic farming, confirming previous results for legume-cereal intercrops. Our study shows that false conclusions can be drawn when analysing intercrops based on only simple indicators and without representing the practical reality. We showed that lentil-spring wheat intercrops could be significantly less profitable than sole cropped lentil when considering the grain yield before mechanical harvest, as profit from spring wheat in intercrop did not economically offset the loss of lentil yield, due to lentil's much higher price. We demonstrated, however, that the presence of spring wheat reduced lentil lodging and allowed a higher percentage of pods to be mechanically harvested. Consequently, after sorting and cleaning grain, the intercrops had significantly higher marketable yield than sole cropped lentil, and led to higher marketable gross margins thus demonstrating that these intercrops can be more profitable. Unfortunately, intercropping did not significantly decrease bruchid damage, which was high in both experimental years. Intercropping can limit risk when yields of sole cropped lentil are low and increase gross margins when they are high.

Our adaptation of the yield gap concept may be used for future studies of legume-cereal intercrops or any other cropping system. The conceptual framework of the yield gap – including a novel definition for mechanically harvested and marketable grain yields – is designed to mimic farmers' real working conditions and thus greatly increase the application potential of scientific results, as farmers can relate the results directly to their practices. The addition of these two reducing factors is particularly relevant for lentil because of its high susceptibility to lodging and bruchid damage and its production as human food. These new production stages complement yield gap analysis and allow for full stepwise quantification of grain losses from attainable yield down to marketable yield. Consequently, farmers would be more likely to adopt more sustainable agricultural practices such as intercropping. Further research is needed, however, particularly to analyse factors that can influence intercrop performances, such as the type of combine harvester and traits of lentil cultivars that can affect mechanical harvest efficiency. From an economic viewpoint, questions remain about how to reduce the cost of grain cleaning and sorting tools, which would increase economic performance of intercrops. Finally, we show that effective biocontrol methods and lentil cultivars tolerant to bruchids are still needed, as bruchids greatly decrease lentil yield in organic farming, especially in areas where they are established, such as southwestern France.

Chapter II. Species niche complementarity for nitrogen improves lentil-wheat intercrop grain production in organic farming

Vigui er Lo ic, Journ et Etienne-Pascal, Justes Eric and Bedoussac Laurent

This chapter is under review in Field Crop Research

Adapted from manuscript



Abstract

Lentil (*Lens culinaris* Med.) is an important component of the human diet in populated areas of the world, but its consumption in Europe remains low despite a recent increasing trend. However, Europe produces only 26% of the lentils it consumes, partly due to strong agronomic constraints that reduce yield, such as bruchid beetles, lodging and weeds.

The objectives of this study were to (1) assess the performance of intercropping lentil with spring wheat to produce lentil under organic farming conditions, (2) evaluate effects of species density and (3) analyse the relationship between nitrogen acquisition and grain attainable yield. A two-year field experiment that followed organic farming rules was carried out in southwestern France in 2015 and 2016. Four lentil cultivars of commercial interest and two spring wheat cultivars were grown as sole crops and in 25 bi-specific intercrops including substitutive and partial additive designs. Mean total intercrop grain yield was higher than the mean yield of sole crops (2.23 ± 0.49 t ha⁻¹ and 1.84 ± 0.38 t ha⁻¹, respectively), similar to that of lentil (2.14 ± 0.75 t ha⁻¹) and higher than that of wheat (1.53 ± 0.10 t ha⁻¹). Similar results were found for the nitrogen (N) accumulated in shoots, which was strongly correlated with the yield for both species and cropping systems. Intercrops had higher yield mainly because of their complementary use of nitrogen pools through the lentil's symbiotic N₂ fixation, which was higher in intercrops than in sole crops ($84 \pm 4\%$ vs. $70 \pm 6\%$ of N acquired, respectively). Increasing wheat density tended to decrease lentil yield and increase wheat yield, which highlights the need to maintain a low wheat density in intercrops. Wheat grain protein concentrations increased in intercrops at the expense of grain yield. We conclude that to maximise lentil yield and the concentration of wheat grain protein, the most effective intercrop is lentil at full density (100% sole crop density) with wheat at low density (15-20% sole crop density). Thus, as for other intercrops, the effectiveness of lentil-wheat intercrops depends on both competition and complementarity between lentil and wheat for available resources, as shown here for nitrogen.

Keywords

Land Equivalent Ratio; Protein; Cultivar; Density; Nitrogen fixation; Nitrogen acquisition

3.1. Introduction

Lentil (*Lens culinaris* Med.) is not widely consumed in Europe, even though it is an important component of the human diet in Asia and Africa. On these continents, lentils and other pulses represent the main source of protein due mainly to their relatively low price and high productivity on dry, marginal lands (CGIAR, 2014; Stefaniak and McPhee, 2015). Lentil consumption is slowly increasing in France and is likely to increase in the future with increased communication about the health benefits of legumes (e.g. FAO 2016 International Year of Pulses). Current Western food systems have contributed to the increase in heart disease, type 2 diabetes and obesity (Duru et al. 2017; Magrini et al. 2016; Polak et al. 2015). In response, plant-based diets are recommended (Duru et al. 2017; Polak et al. 2015). They rely greatly on consuming legume grains to meet protein requirements, as these grains have high protein concentrations along with low carbohydrate and fat contents (Savage, 1991; Chardigny and Walrand, 2016).

At the global scale, about two-thirds of lentil production is consumed in the same area in which it is produced (CGIAR, 2014; Stefaniak and McPhee, 2015). However, the European Union produces only 26% of its lentil consumption (FAOSTATS, 2013), and France produces only 45% of its consumption. In France, this is due to several factors, such as public policies focused on cereal production and market dynamics which are unfavourable to grain legumes (Magrini et al. 2016). Consequently, increasing lentil production is desirable to decrease imports and thus meet demand in France, and more broadly in Europe. This increase is particularly necessary for organic lentil, for which demand is strong and increasing because consumers increasingly prefer more locally and organically produced plant protein.

Like other legumes, lentil has many other advantages, especially agronomic. Cultivating legumes improves in-field biodiversity, which helps break pathogen cycles. It decreases fertiliser use, increases nitrogen (N) availability for the following crop (thereby decreasing the risk of water and air pollution), and limits greenhouse gas emissions (Bedoussac et al. 2015; Duru et al. 2017). These advantages are due mainly to legumes' biological N₂ fixation, due to their roots' symbiotic relationship with rhizobacteria (Büchi et al. 2015; Reda, 2015). Although lentil has a high N requirement (28 mg N g⁻¹ of shoot dry matter vs. 16 mg for wheat; Sinclair and De Wit, 1975; Whitehead et al. 2000), it can meet up to 80% of it from N₂ fixation (Reda, 2015). This ability is particularly useful in low N systems such as stockless organic farming, in

which N often becomes a limiting factor due to the prohibition on using inorganic fertilisers and cost of organic fertilisers.

Unfortunately, many factors can decrease lentil grain yield, such as (1) bruchid beetles (Coleoptera: Bruchidae) whose larvae feed on developing lentil grains (Delobel, 2005; Yus-Ramos et al. 2014), which renders them inedible and causes up to 50% loss in income for farmers (Laserna-Ruiz et al. 2012; Viguier et al. 2018); (2) lodging, which decreases the effectiveness of mechanical harvest and results in in-field grain losses up to 75% (Viguier et al. 2018) and (3) weeds, which can cause grain losses up to 60-100% (Wang, 2013). The combination of these three factors is one reason for the low hectareage of lentils in France and Europe. Farmers are reluctant to grow crops with low and unstable yields, especially in organic farming, for which no effective (bio)control method against bruchids or weeds exists.

Intercropping, simultaneously growing two or more species in the same field for a significant period but without necessarily sowing or harvesting them at the same time (Willey, 1979), seems an interesting solution for lentil production. Several studies have shown promising results of intercropping lentil with cereals, especially when N availability is low, such as in organic farming (Yu et al. 2016). It has been shown to increase total grain yield (Akter et al. 2004; Carr et al. 1995), as well as cereal grain protein concentration (Wang et al. 2013) and gross margin (Akter et al. 2004; Viguier et al. 2018). These results could be due to reduced lodging (Carr et al. 1995; Erskine et al. 1991; Viguier et al. 2018) and fewer weeds (Wang et al. 2013), though no effect on bruchid damage was observed (e.g. Viguier et al. 2018).

Although intercropping lentil with spring wheat appears a suitable agricultural practice, it is not widespread in south-western France, even in organic farming. This is due mainly to the lack of knowledge about optimal species densities in interaction with cultivars of interest, which could be key factors to design innovative intercropping management practices and improve the robustness of intercropping for lentil production. As Yu et al. (2016) indicate, species densities in intercrops influence interspecific interactions and consequently the productivity of crops in mixtures. Little is known about the effects of intercrops on N acquisition by spring crops (i.e. those with short growth periods), though N acquisition has a strong influence on yield as seen in other grain legume-cereal intercrops (Bedoussac et al. 2015).

The objectives of this study, which examined lentil-spring wheat intercrops under organic farming conditions, were to (1) compare the performance of four lentil cultivars of commercial interest intercropped with two spring wheat cultivars to those of their respective sole crops; (2)

evaluate the influence of species density, which governs interspecific competition in intercrops, on yields and (3) analyse N acquisition, grain yield and their relationship to explain the yield of both species in intercrops.

3.2. Materials and Methods

3.2.1. Site, soil, and climate

A two-year field experiment was carried out at the Institut National de la Recherche Agronomique station in Auzeville (south-western France, 43°31'N, 1°30'E) in 2015 and 2016. In 2015, the soil was sandy clay loam (24% clay, 22% silt and 54% sand) with a total soil water content at sowing of 294 mm (0-120 cm soil depth). In 2016, the soil was loam (25% clay, 33% silt and 44% sand) with a total soil water content at sowing of 286 mm (0-120 cm). Soil mineral N at sowing (0-120 cm) was low: 31 and 20 kg N ha⁻¹ in 2015 and 2016, respectively. Degree days over the growing period (base temperature = 0°C) in 2015 (3060°C-days) was higher than its 20-year mean at the experimental site (2901°C-days), but that in 2016 (2766°C-days) was lower than the mean. Mean daily maximum temperature from lentil flowering to maturity (a key period for lentil development) was higher in 2015 than in 2016 (29°C vs. 26°C, respectively). Total rainfall during the entire growing period was similar in both years (305 and 286 mm in 2015 and 2016, respectively) and close to the 20-year mean (282 mm). However, rainfall distribution differed between the two years. In particular, 57% of rainfall in 2015 was concentrated in two storm events (early May and mid-June), without any rainfall in between. These conditions may have generated water stress for plants from mid-May to mid-July. Conversely, rainfall in 2016 was homogeneously distributed throughout the growing period, for which we assumed that no water stress occurred.

3.2.2. Experimental design

The experiment was a randomised block design with three replicates. Four lentil cultivars were used – Anicia, Beluga, Flora, and Rosana, yielding green, black, yellow and red grains, respectively – that differ in precocity and yield potential. Two spring wheat (*Triticum aestivum*, referred to as “wheat” in the text for abbreviation) cultivars – Valbona and Togano – differing in precocity were used. Each cultivar was grown as a sole crop for reference with an emergence objective of 450 plants m⁻² for wheat and 300 plants m⁻² for lentil, corresponding to the density advised by expert knowledge on organic farming in southern France to maximise yield. Twenty-five bi-specific intercrops composed of one lentil cultivar and one wheat cultivar, differing in

cultivars and densities (in substitutive, also referred as replacement in literature and partial additive designs), were evaluated (Table 1). Densities of lentil and wheat ranged from 100% and 17% of the reference, respectively (100L/17W), to 50% and 50% of the reference, respectively (50L/50W).

Table 1 Intercrop and sole crop treatments performed in the field in 2015 and 2016. In each intercrop treatment, each lentil cultivar indicated was grown with each spring wheat cultivar indicated.

Cropping system	Species density (% of the reference)		Cultivars						Total
			Lentil			Wheat			
	Lentil	Wheat	Anicia	Beluga	Flora	Rosana	Togano	Valbona	
Intercrops	100	50	X	X			X	X	n=7*
	100	33	X	X	X	X	X	X	n=16
	100	17	X	X	X	X	X	X	n=16
	67	33	X	X			X	X	n=8
	50	50	X					X	n=2
Sole crops	100	0	X	X	X	X			n=8
	0	100					X	X	n=4

* the Anicia-Valbona treatment was not harvested in 2016 due to a sowing issue, decreasing n by 1

Crops were sown on 12 March 2015 and 23 March 2016. Each plot consisted of 10 rows spaced 16.5 cm apart which were 10 m long in 2015 and 8 m long in 2016. In intercropped plots, the two species were homogeneously mixed within each row. The experiment was conducted under organic farming rules and no pesticides or fertilisers were applied. Weeds were removed from plots when necessary to avoid the confounding influence of a heterogeneous spatial distribution of weeds. Other than bruchid damage to lentils, no significant yield-reducing biotic factors were observed for the sole crops or intercrops.

3.2.3. Sampling and measurements

The number of seedlings in 3 rows 1 m long within the 6 inner rows of each plot was counted one month after emergence to measure plant densities. At lentil maturity, in mid-July in both years, an area of 1.98 m² (6 inner rows 2 m long to avoid edge effects) was hand-harvested. Wheat always reached maturity before lentil, but no wheat grain was lost despite the delay in harvest, due to its indehiscence. Lentil shoots were carefully hand-harvested to prevent pod opening and grain loss. Wheat was cut at ground level with pruning shears. Crops were threshed separately using a research-designed thresher (brand Roland Chateau du Loir, France) to separate grains. Grains were cleaned in an air column to separate heavy grains (sound grains) from light grains (small grains and lentil bruchid-damaged grains). Heavy grains, light grains and wheat and lentil straw were then oven-dried for 48 hours at 80°C to obtain total dry weights

and determine one-grain-weights. Grains in each fraction were counted to determine the number of grains per m², corresponding to sound grains plus bruchid-damaged grains.

For each treatment and each crop, samples of straw and sound grains were ground separately (0.5 mm), and N content was determined using a Dumas combustion method with a LECO-2000 analyser (LECO Corporation, St. Joseph, Michigan, USA). ¹⁵N concentration in vegetative parts and sound grains was assessed using an elemental analyser (Euro-EA, Eurovector, Milan, Italy) and a mass spectrometer (Delta advantage, Thermo-Electron, Bremen, Germany).

3.2.4. Estimate of attainable lentil yield

Bruchid damage and mechanical harvesting cause significant and variable losses of lentil grains which render comparison of actual yields or mechanically harvested yields inaccurate (Viguiet et al. 2018). Therefore, we considered the attainable lentil yield, which is the amount of lentil grain that would be produced in the absence of these factors. We assumed that bruchid larvae damaged developing lentil grains sufficiently late in the crop reproductive phase that lentil plants could not compensate for the damage, and that lentil yield components (i.e. grains per m² and one-grain-weight) were similar to those expected in the absence of bruchids. The attainable lentil yield (t ha⁻¹) of each plot was estimated as grains per ha × one-grain-weight (t). Henceforth, the term “grain yield” refers to “attainable yield”. For wheat, we observed no major reducing factors during crop growth; therefore, hand-harvested grain yield was similar to attainable grain yield.

3.2.5. Calculations and analysis

3.2.5.1. Nitrogen acquisition

The N accumulated by lentil or wheat (kg N ha⁻¹) in grain (QNGrain), straw (QNStraw) and shoots (QNShoots) was calculated as follows:

$$QNGrain = GrainYield \times \%NGrain$$

$$QNStraw = StrawYield \times \%NStraw$$

$$QNShoots = QNGrain + QNStraw$$

where GrainYield is the grain yield (expressed here in kg ha⁻¹), %NGrain is the percentage of N in the grain, StrawYield is the straw yield (kg ha⁻¹) and %NStraw is the percentage of N in the straw.

3.2.5.2. Nitrogen fixation

The percentage of accumulated N which is fixed from the air (%Nfix) was calculated for lentil in intercrops and sole crops using the variation in the natural abundance of ¹⁵N in shoot N. In this method, the ¹⁵N abundance in lentil ($\delta^{15}N_L$) and a non-N₂-fixing reference crop ($\delta^{15}N_{ref}$, corresponding to wheat shoots) is expressed as a difference (δ , in ‰) from the international standard for atmospheric N₂ (0.3663% of ¹⁵N). A correction factor β , corresponding to the $\delta^{15}N$ in lentil shoots, depends completely on N₂ fixation (Unkovich et al. 2008) and was assumed to be equal for all cultivars (-1.5‰), in agreement with Shah et al. (1997). The percentage of N fixed from the air was calculated for lentil according to Shearer and Kohl (1986), as follows:

$$\%Nfix = \frac{\delta^{15}N_{ref} - \delta^{15}N_L}{\delta^{15}N_{ref} - \beta} \times 100$$

We assumed that using wheat for $\delta^{15}N_{ref}$ was a suitable proxy of the $\delta^{15}N$ of the soil mineral N available for lentil (Unkovich et al. 2008; Bedoussac and Justes, 2010). To capture the variability in the $\delta^{15}N$ of soil mineral N, we calculated one value of $\delta^{15}N_{ref}$ for each block as the mean of the two wheat cultivars in intercrops and sole crops. Then, the percentage of N fixed from the air was calculated for lentil in each plot using the $\delta^{15}N_{ref}$ of the same block in the experiment.

The amounts of N fixed from the air (QNfix, kg N ha⁻¹) and of mineral N (QNmin, kg N ha⁻¹) acquired from the soil by lentil in intercrops or sole crops were calculated as follows:

$$QNfix = QNShoots \times \%Nfix$$

$$QNmin = QNShoots - QNfix$$

For wheat, QNmin = QNShoots, as wheat cannot fix N₂ from the air.

3.2.5.3. Land equivalent ratio

The land equivalent ratio (LER) is commonly used to compare the performance of intercropped species to those of their respective sole crops. The LER is the relative land area required when growing sole crops to produce the same yield as the intercrop (with the same species

percentages) or to acquire the same amounts of N as the intercrop (De Wit and Van Den Bergh, 1965). In a bi-specific intercrop, LER is the sum of the partial LER of each species, as follows:

$$LER_L = \frac{X_{L-IC}}{X_{L-SC}}$$

$$LER_W = \frac{X_{W-IC}}{X_{W-SC}}$$

$$LER = LER_L + LER_W$$

where “X” is the grain yield or N accumulated in shoots per unit of surface area for lentil in the intercrop (L-IC), lentil in the sole crop (L-SC), wheat in the intercrop (W-IC) and wheat in the sole crop (W-SC). Partial LER values were calculated per year for each pair of cultivars in each intercrop treatment, using the mean of the three replicates of the respective sole crop. The sole crop reference was standardised in this way to mask the variability due to sole crops, thus introducing only the variability associated with intercrops (Bedoussac and Justes, 2010).

3.2.6. Statistics

Shapiro-Wilk and Levene’s tests were used to test the normality of the data and the homoscedasticity of its variance, respectively. Analysis of variance were performed using the AOV procedure of R software v. 2.7.1 (R development Core Team 2007) and its assumptions were checked by visual examination of the residuals plotted against predicted values. Tukey’s Honest Significant Difference test with $\alpha=0.05$ was used to compare grain yields and nitrogen accumulation between species, cultivars and cropping system. All results are presented as mean \pm 1 standard deviation.

3.3. Results

3.3.1. Grain yield and nitrogen acquisition in total intercrops and mean sole crops

Averaged over both years, total intercrop grain yield was higher than the mean grain yield of sole crops (2.23 ± 0.49 and 1.84 ± 0.38 t ha⁻¹, respectively; $P < 0.01$), similar to that of sole cropped lentil (2.14 ± 0.75 t ha⁻¹, $P < 0.001$), and higher than that of sole cropped wheat (1.53 ± 0.10 t ha⁻¹, $P < 0.001$) (Fig. 8a). Grain yield was higher in 2016 than in 2015 for the total intercrop ($P < 0.001$) and sole cropped lentil ($P < 0.05$), but no difference was observed between years for sole cropped wheat.

Averaged over both years, total intercrop N accumulated in shoots was higher than the mean N accumulated by sole crops (110 ± 23 and 94 ± 20 kg N ha⁻¹, respectively; $P < 0.01$), lower than that of sole cropped lentil (142 ± 42 kg N ha⁻¹; $P < 0.001$), and much higher than that of sole cropped wheat (47 ± 2 kg N ha⁻¹; $P < 0.001$), (Fig. 8b). N accumulated in shoots was higher in 2016 than in 2015 for sole cropped lentil ($P < 0.05$), but no difference was observed between years for the total intercrop or sole cropped wheat. For both grain yield and N accumulated in shoots, the higher the mean value for the sole crop, the higher the mean value for the total intercrop (Figs. 8a and 8b).

No significant difference was observed in the amount of mineral N acquired between the total intercrop, sole cropped lentil and sole cropped wheat (46 ± 7 , 37 ± 10 and 47 ± 2 kg N ha⁻¹, respectively), which indicates that each cropping treatment was able to acquire all the mineral N available from the soil. Lentil in intercrops acquired less mineral N than sole cropped lentil (12 ± 5 vs. 37 ± 10 kg N ha⁻¹, respectively, $P < 0.001$). No difference in the amount of mineral N acquired was observed between years for all species and cropping treatments, except for wheat in intercrops, which acquired more mineral N in 2016 than in 2015 (38 ± 5 vs. 29 ± 5 kg N ha⁻¹, respectively; $P < 0.001$).

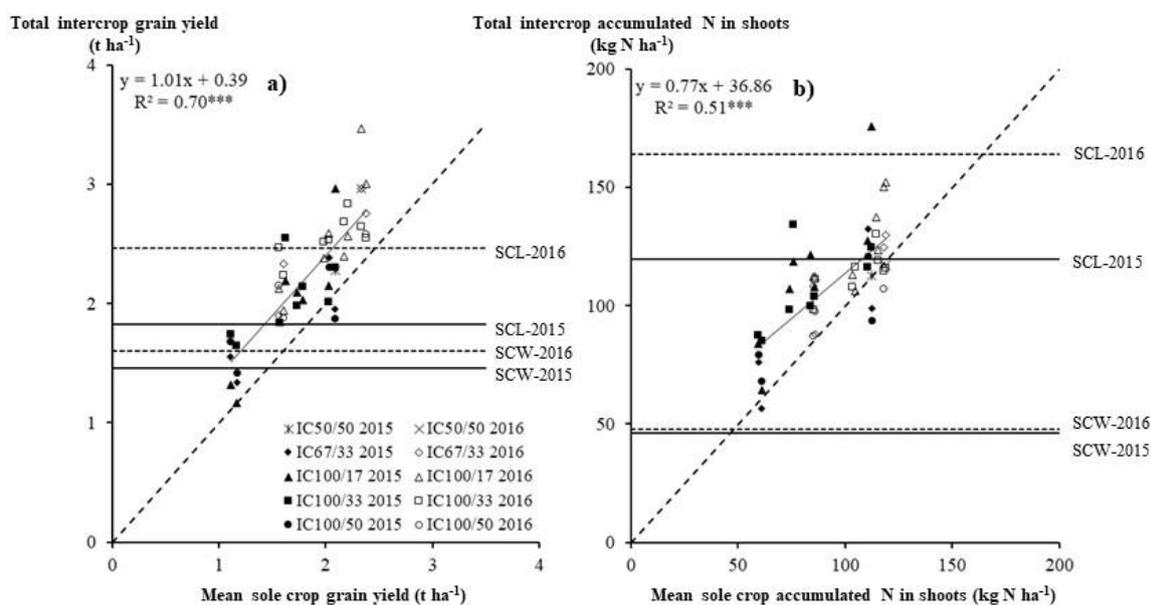


Fig. 8 (a) Grain yield and (b) nitrogen accumulated in shoots of the total intercrop (IC) and sole crops (SC). *** indicates $P < 0.001$; $n = 49$. Symbols indicate relative (%) densities of lentil/wheat and the experimental year. Solid and dashed horizontal lines indicate mean values for spring wheat in SC (SCW; both cultivars combined) and of lentil in SC (SCL; all cultivars combined) in 2015 and 2016, respectively. The same symbols are used in both figures.

3.3.2. Species density drives competition in intercrops

Lentil grain yield was higher ($P < 0.05$) in 100L/17W than in 100L/50W and was similar to that in 100L/33W, except for Anicia (Table 2). Conversely, wheat grain yield in intercrops tended to increase as wheat density increased, except for Flora (Table 2). For a given wheat density, decreasing lentil density from 100% to 67% or 50% had no effect on grain yield of lentil or wheat. In both 67L/33W and 50L/50W, however, grain yield of lentil was lower than that in 100L/17W (Table 2). In 100L/17W, grain yield of Anicia ($1.90 \pm 0.38 \text{ t ha}^{-1}$; $P < 0.05$) was the highest and that of Beluga ($0.91 \pm 0.34 \text{ t ha}^{-1}$; $P < 0.05$) was the lowest, while those of Flora and Rosana (1.36 ± 0.16 and 1.47 ± 0.01 , respectively) were intermediate and similar.

Table 2 Effect of species densities and lentil cultivar on intercropped lentil and wheat grain yields averaged over both experimental years. Individual pairwise comparisons were performed between the density of each lentil cultivar and those of the averaged, associated wheat cultivars. Absence of common letters denotes statistically significant difference in means ($P<0.05$).

Species density (% of the reference)		Lentil cultivar							
		Anicia		Beluga		Flora		Rosana	
Lentil	Wheat	Lentil	Wheat	Lentil	Wheat	Lentil	Wheat	Lentil	Wheat
100	50	1.02±0.16 ^c	0.82±0.13 ^{ab}	0.62±0.20 ^b	0.78±0.09 ^b				
100	33	1.33±0.12 ^b	0.68±0.10 ^{ab}	0.85±0.21 ^a	0.77±0.14 ^b	1.26±0.31 ^a	0.68±0.14 ^a	1.37±0.16 ^a	0.71±0.17 ^b
100	17	1.90±0.38 ^a	0.64±0.13 ^b	0.91±0.34 ^a	0.47±0.07 ^a	1.36±0.16 ^a	0.52±0.13 ^a	1.47±0.01 ^a	0.57±0.21 ^a
67	33	1.33±0.30 ^{bc}	0.77±0.13 ^{ab}	0.62±0.25 ^b	0.75±0.09 ^b				
50	50	1.23±0.12 ^{bc}	0.91±0.22 ^a						

3.3.3. Land equivalent ratios for grain yield and nitrogen accumulated in shoots

LERs calculated from grain yield (Fig. 9a) or the N accumulated in shoots (Fig. 9b) were always similar to or higher than 1, with no significant difference between years. All but one partial LER of lentil (LER_L) was lower than 1, indicating that lentil in intercrops, compared to sole cropped lentil, had lower grain yield ($1.18±0.41$ vs. $2.14±0.75$ t ha⁻¹, respectively; $P<0.05$) and accumulated less N in shoots ($76±22$ vs. $142±42$ kg N ha⁻¹, respectively; $P<0.001$), when averaged over both years. Likewise, all but one partial LER of wheat (LER_W) was lower than 1, indicating that wheat in intercrops, compared to sole cropped wheat, also had lower grain yield ($1.06±0.28$ vs. $1.53±0.10$ t ha⁻¹, respectively; $P<0.01$) and accumulated less N in shoots ($33±7$ vs. $47±2$ kg N ha⁻¹, respectively; $P<0.01$), when averaged over both years.

Linear regressions indicated that for both grain yield and N accumulated in shoots, the higher the LER_W , the lower the LER_L (Fig. 9a and 9b). In addition, for both grain yield and N accumulated in shoots, as wheat density increased, LER_L decreased and LER_W increased. On average, LER_L calculated from grain yield or N accumulated in shoots was higher in 2015 than in 2016, while the opposite was observed for LER_W .

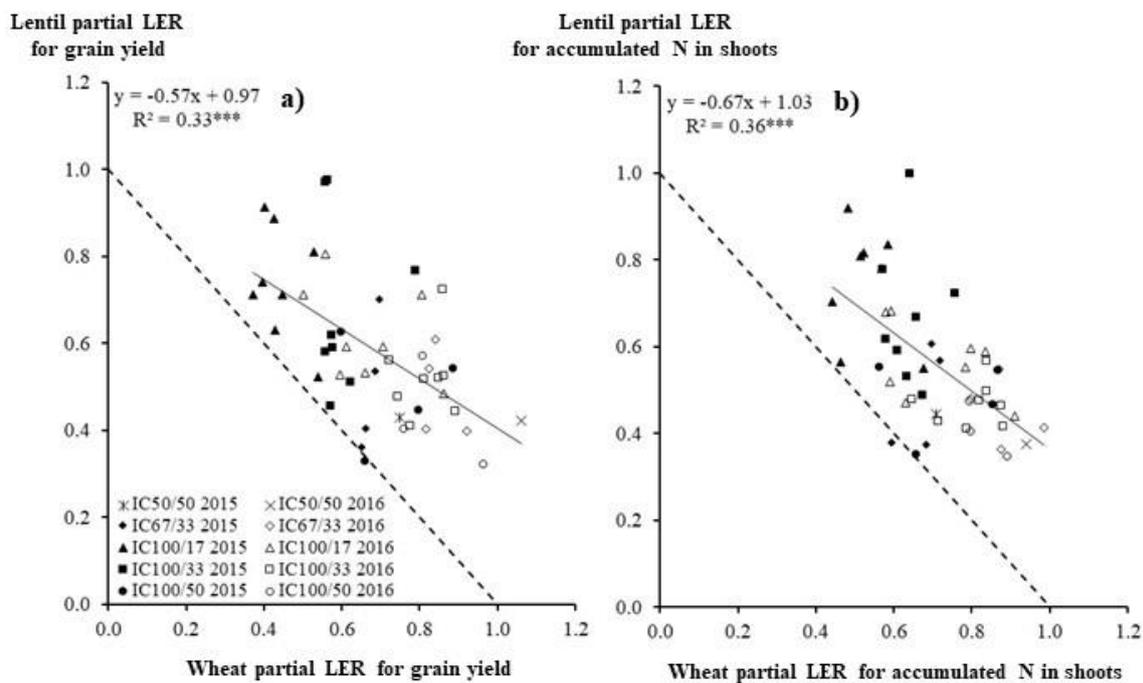


Fig. 9 Partial land equivalent ratio of lentil in intercrops (LER_L) vs. partial land equivalent ratio of wheat in intercrops (LER_W) calculated from (a) grain yield or (b) N accumulated in shoots. *** indicates $P < 0.001$; $n = 49$. Symbols indicate relative (%) densities of lentil/wheat and the experimental year. Dashed lines correspond to $LER = 1$.

3.3.4. Grain yield is strongly correlated with nitrogen accumulated in shoots

For both wheat and lentil, grain yield was strongly correlated with N accumulated in shoots (Figs. 10a and 10b). The species differed in N requirements, however, with wheat needing ca. half as much N as lentil to produce an equivalent grain yield.

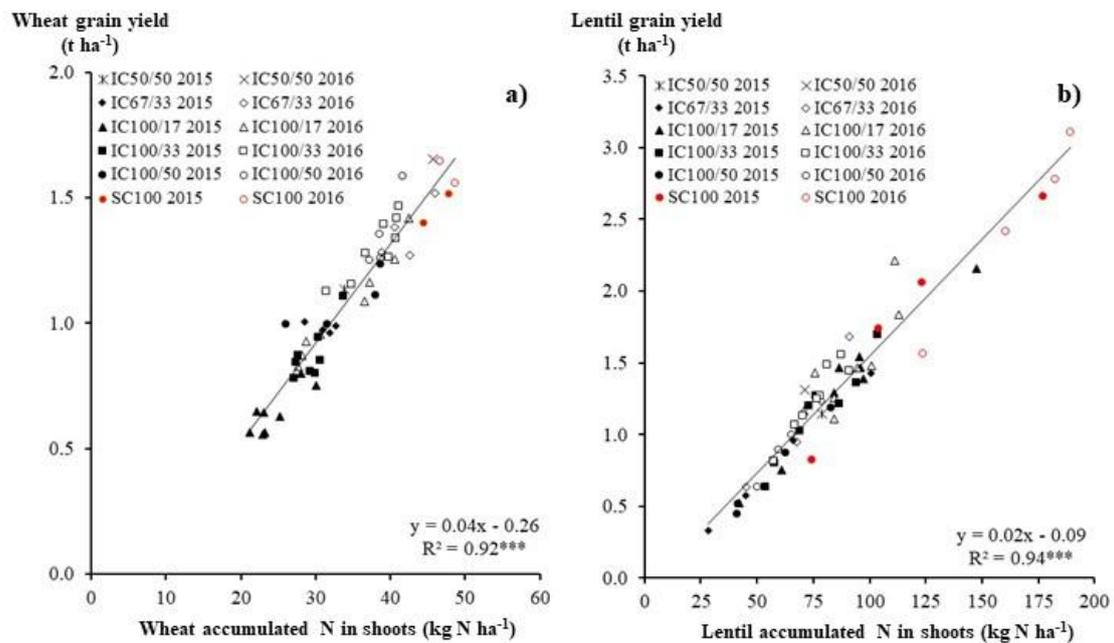


Fig. 10 Grain yield vs. nitrogen accumulated in shoots for (a) wheat and (b) lentil in intercrops (IC) and sole crops (SC). *** indicates $P < 0.001$; $n = 49$. Symbols indicate relative (%) densities of lentil/wheat, the experimental year and cropping treatment.

3.3.5. Lentil fixes a higher percentage of nitrogen in intercrops but in lower amounts

For lentil, the intercrop:sole crop ratio of the percentage of accumulated N fixed from the air always exceeded 1.0 (Fig. 11), indicating a higher percentage in intercrops than in sole crops ($84 \pm 4\%$ vs. $70 \pm 6\%$ respectively; $P < 0.001$). Conversely, the intercrop:sole crop ratio of the amount of N fixed from the air was always lower than 1.0, indicating less N₂ fixed in intercrops than in sole crops (64 ± 18 vs. 101 ± 35 kg N ha⁻¹ respectively; $P < 0.001$). Linear regression indicated that the ratio of the amount of N fixed increased as the ratio of the percentage of N fixed increased (Fig. 11). Both ratios were lower in 2016 than in 2015, due to the lower percentage and amount of N fixed in lentil shoots in sole crops in 2015 than in 2016 ($P < 0.05$), while no difference was observed in intercrops.

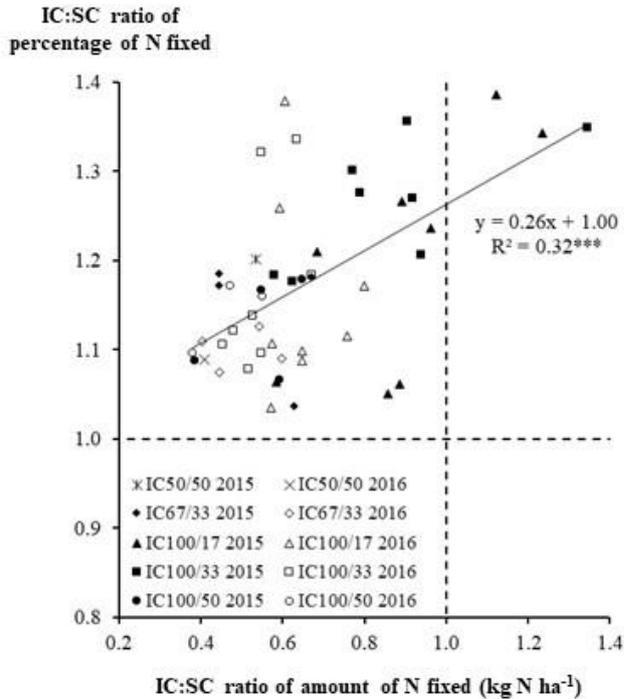


Fig. 11 For lentil, the intercrop (IC):sole crop (SC) ratio of the percentage of nitrogen (N) fixed from the air (%) vs. the IC:SC ratio of the amount of N fixed (kg N ha^{-1}). *** indicates $P < 0.001$; $n = 49$. Symbols indicate relative (%) densities of lentil/wheat and the experimental year. Dashed lines indicate ratios of 1.0 (IC=SC).

3.3.6. Protein concentration is correlated with grain yield for wheat but not for lentil

Linear regression indicated that wheat's grain protein concentration decreased as its grain yield increased ($y = -0.01x + 0.14$; $R^2 = 0.22$, $P < 0.001$; Fig. 12a) and, averaged over all cultivars and years, was higher in intercrops than in sole crops ($12.9 \pm 0.8\%$ vs. $12.0 \pm 0.8\%$; $P < 0.01$). In contrast, in 2016 only, lentil's grain protein concentration increased as its grain yield increased, averaged over all cultivars ($y = 0.01x + 0.24$; $R^2 = 0.45$, $P < 0.001$; Fig. 12b), though no significant difference in mean grain protein concentration was observed between cultivars. In 2015, lentil grain protein concentrations were higher than in 2016 ($P < 0.001$), and no correlation between them and grain yield was observed, when averaged over all cultivars (Fig. 12b). Unlike in 2016, however, lentil cultivars differed in mean grain protein concentration in 2015, with Anicia having a higher concentration than Flora ($34.9 \pm 0.2\%$ vs. $31.1 \pm 0.9\%$, respectively; $P < 0.001$), which itself had a higher concentration than Rosana and Beluga ($27.2 \pm 0.5\%$ and $25.4 \pm 0.5\%$, respectively; $P < 0.001$).

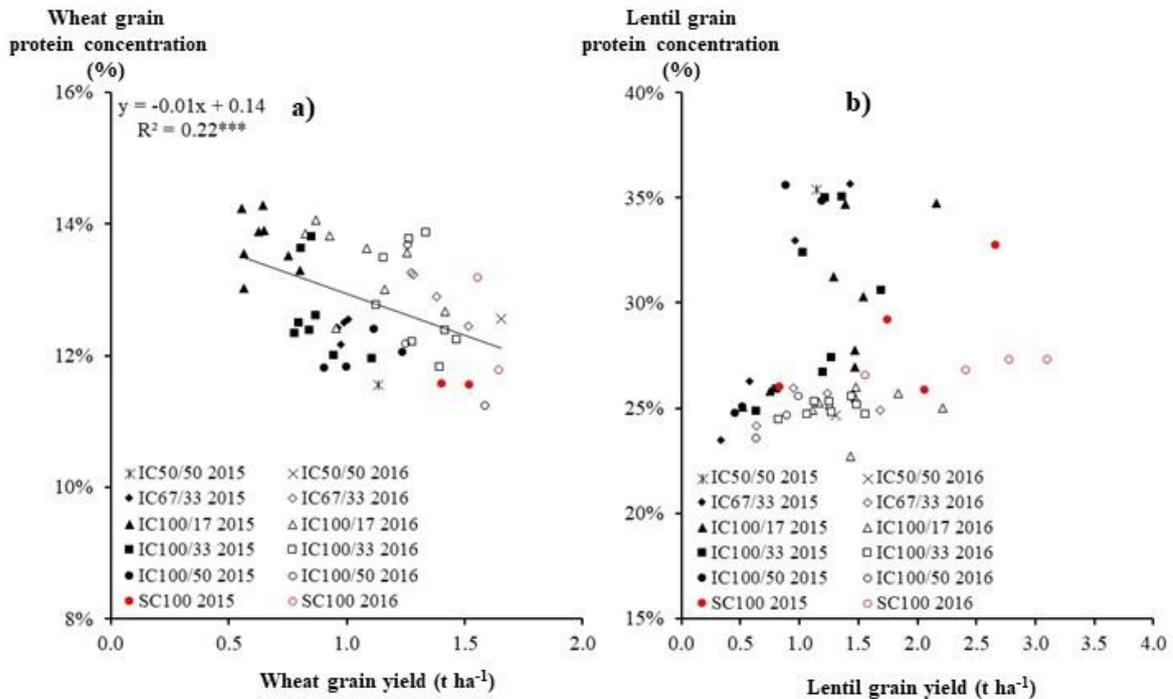


Fig. 12 Grain protein concentrations of (a) wheat and (b) lentil in intercrops (IC) and sole crops (SC) as a function of grain yield. *** indicates $P < 0.001$; $n = 49$. Symbols indicate relative (%) densities of lentil/wheat, the experimental year and cropping treatment.

3.4. Discussion

3.4.1. Grain yield and nitrogen accumulated in the total intercrop vs. sole crops

Soil mineral N at sowing in our field experiment was low and equal in both years, as indicated by the amount of mineral N acquired by sole cropped wheat. When averaged over all treatments, cultivars and years, total intercrop grain yield was 21% higher than the mean grain yield of sole crops (Fig. 8a). This confirms that intercropping lentil with wheat is an effective practice for increasing grain production in low N systems such as stockless organic farming. This increase in yield was also observed in other lentil-wheat intercrops (Akter et al. 2004; Carr et al. 1995) and other legume-cereal intercrops whose production under organic farming conditions was 20-30% higher than that in sole crops (Bedoussac et al. 2015; Hauggaard-Nielsen et al. 2009).

The advantage of lentil-wheat intercrops for total grain production can be explained by the larger amount of N accumulated in shoots of intercrops than the mean amount in sole crops (Fig. 8b), as observed by Whitehead et al. (2000). For sole crops, grain yield in intercrops was strongly related to the amount of N acquired (Figs. 10a and 10b). No difference in the amount of mineral N accumulated in shoots was observed between intercrops and sole crops, indicating

that wheat, lentil and their intercrops had similar abilities to acquire all the mineral N available from soil supplies under our conditions. This suggests that the higher yield of intercrops is due mainly to the niche complementary for N made possible by lentil N₂ fixation and the higher percentage of accumulated N which is fixed in intercrops (Fig. 11) than in sole crops, a result also observed for pea-oat intercrops (Rauber et al. 2001).

3.4.2. Grain yield of and nitrogen accumulated by intercropped species

Grain yield and N accumulated in shoots of each species were lower in intercrops than in sole crops (Figs. 9a and 9b), and this was observed for lentil even when its density was similar in both cropping treatments. This decrease is due to wheat competition for resources (e.g. light, water, mineral N), which limits their availability for lentil and results in decreased biomass and photosynthetic capacity of lentil (Whitehead et al. 2000). In addition, lentil grain yield differed significantly between years in both intercrops and sole crops. Erskine and El Ashkar (1993) reported that differences in seasonal rainfall could explain up to 80% of the variability in lentil grain yield. Compared to the favourable rainfall distribution in 2016, the dry spring in 2015 created water stress during the grain filling period. This explains, at least partially, the difference in yield between the two years. The higher grain yield in 2016 than in 2015 may be due not only to favourable rainfall distribution but also to cooler temperatures in the May of 2016, which correspond better to lentil's physiological requirements (McKenzie and Hill, 2004).

For wheat, grain yield and N accumulated in shoots per plant (data not shown) were higher in intercrops than in sole crops, a result also observed for barley-pea intercrops (Hauggaard-Nielsen et al. 2001). We can assume that wheat grows faster than lentil as both its shoots and roots have an advantage over those of lentil for resource acquisition (e.g. mineral N). This assumption is supported, at least for roots, because wheat in intercrops accumulated significantly more mineral N in both years than lentil in intercrops. As the wheat in intercrops strongly decreased lentil's access to mineral N, the lentil had to rely more on N₂ fixation, as illustrated by the higher percentage of N fixed from the air in intercrops (Fig. 11) than in sole crops (Fig. 11). Therefore, wheat in intercrops had access to a similar amount of soil mineral N as wheat in sole crops, but with a lower plant density, which led to more N available per plant in intercrops than in sole crops.

As previous studies on legume-cereal intercrops have indicated (Naudin et al. 2009; Bedoussac et al. 2015), the increase in the percentage of accumulated N which was fixed in shoots did not

result in a larger amount of N per ha being fixed in shoots (Fig. 11). This is due to wheat competition for abiotic resources (light, water, and nutrients), which decreased lentil shoot biomass by 43% and consequently decreased lentil grain yield – as biomass and grain yield are strongly correlated (Whitehead et al. 2000) – when intercropped in partial additive treatments and averaged over all cultivars and years.

Our results also highlighted the weak correlation between LER_L and LER_W , whether calculated from grain yield or N accumulated in shoots. This result differs from that of Akter et al. (2004), who found a negative correlation between lentil and wheat grain yields in intercrops. These latter results were obtained with fertilisers applied at sowing, which increased cereal biomass and possibly the magnitude of wheat competition. Under our conditions, wheat growth, and therefore its competition with lentil, was limited by the low mineral N available in both years and a relatively low wheat density in intercrops. Even with these limitations, wheat competes strongly with lentil, suggesting that mineral N uptake by the cereal decreased the legume grain yield, a result also observed for other legume-cereal intercrops (Bedoussac et al. 2015). This result confirms that intercropping lentil and wheat is a suitable practice for low N input systems such as stockless organic farming, but only when wheat competition is minimised.

3.4.3. Grain protein concentrations of species in intercrops and sole crops

Grain protein concentration of wheat in intercrops increased about 1% compared to that in sole crops, indicating that more N was available per plant in the former, as previously discussed. The highest mean wheat grain protein concentrations ($13.5 \pm 0.5\%$) were obtained in 100L/17W treatments, in which wheat density and grain yield were the lowest (Table 2). This increase in wheat grain protein concentration is due mainly to wheat's lower grain yield when intercropped (Fig. 12a) and because the lentil fixes a high percentage its N from the air (Fig. 11), indicating that lentil leaves most of the soil mineral N available for wheat. This increase in cereal grain protein concentration was also observed in barley-lentil intercrops (Wang et al. 2013) and other legume-cereal intercrops (Gooding et al. 2007; Naudin et al. 2009; Bedoussac et al. 2015).

Cereal grain with high protein concentration is likely to have a premium price, which contributes to the economic advantages of intercrops vs. sole crops, especially in organic farming, in which it is difficult to obtain high grain protein concentrations due to the prohibition on using inorganic fertilisers and the low efficiency of organic fertilisers. As producing such a high protein concentration in wheat grain in organic agriculture is challenging for farmers, it could become a production objective *per se*, along with producing lentils in intercrops. This

result is another reason to encourage intercropping. Nevertheless, an economic assessment of intercrops is needed to identify the best compromise, due to the negative correlations between (1) wheat grain yield and protein concentration and (2) lentil grain yield and wheat grain yield. No significant difference was found in grain protein concentration for lentil in intercrops and sole crops (Fig. 12b), which is consistent with the studies of Wang et al. (2013) and Burstin et al. (2011). For both intercrops and sole crops, lower lentil grain protein concentration in 2016 than in 2015 could be due to the higher yield in 2016 than in 2015 ($P < 0.05$). Hamdi et al. (1991) observed a weak negative correlation between grain protein concentration and grain yield in lentil. Monti et al. (1983) reported that this correlation was found in certain legume species, such as peas and dry beans, but not in others, such as faba beans.

Lentil grain protein concentration ranged from 22.7-35.7% among the four cultivars tested, which agrees with results of Savage (1991), who reported concentrations of 20-36%. Our results suggest that the influence of the environment on lentil grain protein concentration is important, as reported by Burstin et al. (2011). In 2016, grain protein concentration did not differ among cultivars but was positively correlated with grain yield. In 2015, however, it did differ among cultivars but had no correlation with grain yield. This result suggests that an interaction occurred between grain yield, grain protein concentration, cultivar and year (Burstin et al. (2011). Specifically, the cultivar effect may have disappeared in 2016 because high yields decreased grain protein concentrations of all cultivars, while the low yields in 2015 may have allowed expression of cultivar differences.

3.4.4. Strategies to improve lentil production in intercrops

Intercropping lentil appears to be a disadvantageous agronomic practice as it produces lower grain yields than sole cropped lentil. However, this conclusion is based on the attainable grain yield. As shown by Viguier et al. (2018a), lentil lodging was much lower in intercrops than in sole crops (15% vs. 40%, respectively), which increased the efficiency of mechanical lentil harvest (75% vs. 50%, respectively). This led to a similar mechanically harvested yield of lentil in intercrops and sole crops (0.80 t ha^{-1}) but a higher gross margin in intercrops than in sole crops (974 ± 376 vs. $713 \pm 348 \text{ € ha}^{-1}$, respectively) due to the addition of marketable wheat yield. Gross margins of lentil-wheat intercrops depend mainly on the yield of lentil grain, due to its higher price (ca. 4 times as high). Therefore, to maximise lentil yield in intercrops and thus the gross margin, wheat density at sowing must remain low: ca. 15-20% (Table 2).

The decrease in wheat density from 50% to 33% and 17% significantly increased lentil grain yield (Table 2). Restricting wheat density to ca. 15-20% at sowing allows lentil to acquire more N and produce more biomass, remobilising even more assimilates to grain during grain filling and maturation. Akter et al. (2004) also observed this negative correlation between wheat density and lentil grain yield in intercrops. For all types of intercrops, partial additive designs (i.e. lentil sown at the same density as in a sole crop) seem especially useful for lentil because they decrease competition from wheat (Table 2). We recommend, however, not sowing lentils at more than 100% of the sole crop density in intercrops, as doing so did not increase grain yield but increased sowing cost due to the relatively high cost of lentil seeds. This conclusion is based on a lentil-wheat intercrop experiment performed in 2016 under the same conditions, in which 133L/17W treatments with Anicia or Beluga were compared with 100L/17W. No difference in Beluga and wheat grain yields were observed, and that of Anicia was even lower ($P < 0.05$). Finally, even in the 100L/17W treatment, lentil experienced decreased grain yield, N accumulated in shoots and N from N_2 fixation. Therefore, it is also essential to decrease the amount of mineral N available in the soil to decrease wheat competition, especially during tillering, in early plant development.

3.5. Conclusions

The effectiveness of intercropping and species functioning depend on competition and niche complementarity between species for available resources, as shown for the crucial resource N. Intercropping lentil with spring wheat is one way to increase total grain production, due mainly to complementary use of N pools made possible through lentil's ability to fix N_2 , which partially but not completely offsets effects of wheat competition. Wheat production and its high grain protein concentration come at the expense of lentil production, which accumulated less N in intercrops than in sole crops due to strong competition with wheat for soil mineral N and light. To increase lentil production in south-western France, intercropping lentil with spring wheat initially appears to be an inappropriate agronomic practice to encourage. However, lodging of lentil can dramatically reduce its grain yield during mechanical harvest. Consequently, intercropping decreases losses caused by lodging to the point that it becomes relevant for lentil production. Also, the addition of high-quality wheat production under conditions of low N input suggest that intercropping is a valuable and profitable agronomic practice for organic and even conventional farms as long as soil mineral N remains low. From our observations, the most effective intercrop mixing ratio to produce a high lentil yield and wheat with high protein concentration is a partial additive design with lentil sown at its sole crop density and wheat

sown at an optimal low density (15-20% of that in sole crop). This approach aims to provide satisfactory lodging and weed suppression while maximising lentil production and the gross margin of the entire intercrop.

Chapter III. Yield component analysis reveals that lentil-spring wheat intercrop grain yield is determined early during the vegetative stages

Vigui r Lo c, Bedoussac Laurent, Justes Eric and Journet Etienne-Pascal

This chapter is to be submitted

Adapted from manuscript



Abstract

Lentil (*Lens culinaris* Med.) is a grain legume whose production within the European Union corresponds to only 26% of the consumption. It has been overlooked by farmers because of low and unstable yields, notably due to lodging, weeds, and bruchid beetles particularly in organic farming. Therefore, intercropping lentil and spring wheat is considered as option to reduce the impacts of those major agronomical bottlenecks.

However, knowledge to design strategies for lentil production when intercropped with wheat is lacking and experimentation based on sequential measurements of plant growth is costly and time consuming despite its relevance for analysing the intercrop functioning. This study aimed at understanding the dynamics of plant-plant interactions by analysing the effects of environment, wheat density and genotypes on species yield components as a basis for designing optimal intercrop managements.

A two-year field experiment following organic farming rules was carried out in southwestern France in 2015 and 2016. Four lentil and two spring wheat cultivars were grown as sole crops and in partial additive intercrops with lentil (the main cash crop produced) sown at sole crop density while differing in wheat densities (companion species for reducing lentil weaknesses).

Total intercrop grain yield was 21% higher than the mean of sole crops. However, both species grain yields were lower in intercrop than in their respective sole crops due to interspecific interactions (-39% and -13% for lentil and wheat, respectively). The interspecific interactions had significant effects on yield components, slightly lowering plant emergence (-4% for lentil and -6% for wheat) but mostly reducing the number of branches per plant of lentil (-32%) and the number of ears per plant of wheat (-9%), which were the two yield components explaining the final performances. This result indicates that the wheat competition on lentil occurred early during vegetative stages and was quite strong even if wheat was sown at low density (75 plants/m²). Year, wheat density and genotypes had also a significant effect on the intensity of interspecific interactions.

Finally, our work provided novel information that will help to design optimized lentil-spring wheat intercrops, which may help to foster the development of this practice by farmers in order to maximize lentil grain yield in organic farming in France.

Keywords

Interspecific interactions; Competition; Environment; Cultivar; Density;

4.1. Introduction

Lentil (*Lens culinaris* Med.) is a grain legume whose consumption in Europe is low but is currently increasing with communication on health benefits of legumes as part of a plant-based diet (e.g. FAO 2016 international year of pulses). However, lentil production within the European Union is even lower, corresponding to only 26% of its consumption and 45% in the case of France (FAOSTATS, 2018). Lentil production deficit in France is explained by the combination of (1) unfavourable public policies, (2) unfavourable market dynamics and (3) numerous major agronomic constraints (Magrini et al. 2016).

Farmers are reluctant to grow grain legumes and in particular lentil due to low and unstable yields, notably in organic farming where no effective biocontrol method exists against pests and weeds. Lentil yield is reduced by (1) bruchid beetles (Coleoptera: Bruchidae), whose larvae feed on developing lentil grains causing up to 50% income loss for farmers (Laserna-Ruiz et al. 2012; Viguier et al. 2018a), (2) lodging, that limits the mechanical harvest efficiency resulting up to 75% grain losses on field (Viguier et al. 2018a) and (3) weeds which can cause yield losses up to 60%-100% (Wang et al. 2013). However, as for other legumes, lentil has also agronomic advantages such as enhancement of biodiversity that breaks pathogens cycles, reduction of fertilizers use and improvement of nitrogen availability for the following crop thanks to nitrogen fixation and reduction of water and air pollution risk (Bedoussac et al. 2015; Reckling et al. 2016). Together, these advantages participate to an improved sustainability of cropping systems (Schneider et al. 2015; Watson et al. 2017).

Intercropping, the simultaneous growth of two or more species in the same field (Willey 1979) appears as an interesting solution to develop lentil production notably in southwestern France and especially for organic systems. Several studies have shown promising results of intercropping lentil with cereals, notably when nitrogen availability is low such as in organic farming without livestock (Yu et al. 2016). Intercropping lentil with cereals has been shown to enhance the total grain yield (Akter et al. 2004; Carr et al. 1995), cereal grain protein concentration (Wang et al. 2013) and gross margin (Akter et al. 2004; Viguier et al. 2018a). These results are partly explained by a reduced lodging (Erskine et al. 1991; Carr et al. 1995; Viguier et al. 2018a) and fewer weeds (Wang et al. 2013) while no effect has been reported on bruchid damages (e.g. Viguier et al. 2018a). Furthermore, intercropping has been shown to increase yield stability of legume-cereals intercrops over both cereal and grain legumes pure stands (Raseduzzaman and Jensen, 2017), which is particularly important for organic farmers

as they need to secure incomes. In southwestern France, lentil is grown as a spring crop and because of pods dehiscence, the synchronicity of the maturity period is considered as decisive criteria for choosing its companion crop in intercrop. In this context, spring wheat appears as a suitable candidate since its physiological maturity is roughly concomitant with that of lentil and it has an economical interest for production of flour with high protein concentration in organic farming conditions (Viguier et al. 2018a).

Lentil develops slowly at early growth stages which lowers its relative competitive ability towards others species and usually results in lower yields in intercrops than in sole crops (Akter et al. 2004; Wang et al. 2012; Viguier et al. 2018b). Understanding the time-course species interactions and thus yield difference between intercrops and sole crops is essential to choose adapted cultivars and to optimize species densities in intercrop, known to be key features in yield determination (Brooker et al. 2014; Neugschwandtner and Kaul 2014). Intercropping studies must ideally implement sequential measurements of crop growth to better understand the dynamics and mechanisms of competitive interactions (Connolly et al. 1990). However, these analyses are costly and time consuming. Species grain yield can be decomposed into a succession of interdependent and consecutive grain yield components (Bedoussac et al. submitted) established at successive or slightly overlapping developmental stages (e.g. Slafer and Rawson 1994). Therefore, the post-harvest analysis of the formation of yield components could be a simpler but relevant alternative approach to sequential measurements of crop growth.

Yield components of lentil and cereals in sole crop have been extensively studied worldwide and it is well known that variation in yield components depends mainly on crop management (Iverson et al. 2014), environment (Subira et al. 2015; Erskine and Goodrich 1988) and genotype (Ellen 1993; Erskine and Goodrich, 1988). However, very few studies have investigated to what extent yield components of lentils and cereals are affected by intercropping (Monti et al. 2016). Thus, the objectives of the present study are to understand dynamics of plant-plant interactions in intercrops by analysing (1) yield formation of both species and their interaction, as explained by yield components, (2) the effect of environment, wheat density and genotypes to finally propose (3) guidelines to design efficient management strategies for lentil production in intercrop with wheat. To do so, we studied the performances of four lentil cultivars intercropped with two spring wheat cultivars in field experiments carried out in two contrasted years in terms of rainfall distribution and temperatures.

4.2. Material and methods

4.2.1. Site, soil, and climate

A two-year field experiment was carried out at the Institut National de la Recherche Agronomique station in Auzeville –south-western France, 43°31'N, 1°30'E– in 2015 and 2016. Soil was sandy clay loam (25% clay, 23% silt and 52% sand) in 2015 and loam (30% clay, 30% silt and 40% sand) in 2016. Total soil water content at sowing was of 294 mm and 286 mm (0-120 cm soil depth) in 2015 and 2016, respectively. Soil mineral N at sowing (0-120 cm) was low and can be considered as very similar between treatments and years, with 31 and 20 kg N ha⁻¹ in 2015 and 2016, respectively (cf. Viguier et al. 2018b for details) . The sum of daily mean temperatures over the growing period was 2901 °C day and 2766 °C day (on a 0 °C basis) in 2015 and 2016, respectively compared to 3060 °C day for the 20-year mean of the experimental site. Moreover, mean daily maximum temperature from lentil flowering to its maturity (a key period for lentil development) was higher in 2015 than in 2016 (29°C vs. 26°C, respectively). Total rainfall during the whole growing period was similar in both years (305 and 286 mm in 2015 and 2016, respectively) and similar to the 20-year mean (282 mm). However, rainfall distribution was contrasted during the two years. In particular, 57% of rainfall was concentrated in two storm events in 2015 (early May and mid-June), without any rain in between. Conversely, rainfall in 2016 had homogenous distribution throughout the growing period. These conditions may have led to water stress for plants from mid-May to mid-July in 2015 while we assumed that no water stress occurred in 2016.

4.2.2. Cultivars and densities

Lentil (*Lens culinaris*) and spring wheat (*Triticum aestivum*) were grown as 3 bi-specific intercrops (IC) in partial additive designs all with lentil sown at sole crop density but differing in wheat densities: IC100%L:17%W, IC100%L:33%W and IC100%L:50%W. The partial additive design was chosen to favour the yield of lentil since it is the most profitable crop. The wheat is used as a companion species mainly to reduce lodging and improve lentil harvest efficiency. It also generates a complement of revenue for farmers after grain sorting (Viguier et al. 2018a). Intercrops were compared to their respective reference sole crops (SC), with an objective of 300 plants m⁻² for lentil (SC100%L) and 450 plants m⁻² for wheat (SC100%W) as advised by expert knowledge to maximize grain potential on organic farming in south-western France. For wheat, sole crops densities of 150 and 225 plants m⁻² corresponding to 33% and 50% of the reference plant density (SC33%W and SC50%W, respectively) were also used. This

allows the evaluation of interspecific interactions in the IC100%L:33%W and IC100%L:50%W treatments respectively. Note that the SC17%W was not performed in field and therefore, interspecific interactions in the IC100%L:17%W were not analysed. Lentils cultivars were Flora, Anicia, Rosana and Beluga yielding yellow, green, red and black grains respectively and ordered by precocity (time from emergence to flowering) from the earliest to the latest. Wheat cultivars were Valbona and Togano, also ordered by precocity. All combinations of densities and cultivars evaluated in both sole crops and intercrops are detailed in Table 3.

Table 3 Field treatments performed in 2015 and 2016. In each intercrop (IC) treatment, lentil was intercropped with each wheat cultivar separately. The asterisk informs that modality IC100%L:50%W with Anicia and Valbona was not available in 2016 due to sowing issue resulting in n=7 instead of n=8 for this modality.

Cropping system	Name of treatments	Species density (% of the SC reference)		Lentil cultivars				Spring-wheat cultivars		Total number of treatments over the two years
		Lentil (300 plants m ⁻²)	Wheat (450 plants m ⁻²)	Anicia	Beluga	Flora	Rosana	Togano	Valbona	
IC	IC100%L:17%W	100	17	X	X	X	X	X	X	n=16
	IC100%L:33%W	100	33	X	X	X	X	X	X	n=16
	IC100%L:50%W	100	50	X	X			X	X	n=7*
SC	SC100%L	100	0	X	X	X	X			n=8
	SC33%W	0	33					X	X	n=4
	SC50%W	0	50					X	X	n=4
	SC100%W	0	100					X	X	n=4

4.2.3. Experimental design and crop sampling

Crops were sown on 12 and 23 March in 2015 and 2016, respectively. Each plot consisted of 10 rows spaced 16.5 cm apart, and of 10 m and 8 m long in 2015 and 2016, respectively. In intercrops, the two species were homogeneously mixed within each row. The experiment was a randomized block design with three replicates. The experiment was conducted without synthetic pesticides or chemical fertilizers. Weeds were handily removed from plots two times during crop growth. Besides bruchids damages on lentil, no other significant yield-reducing biotic factor were observed on any of the treatments and for both species. At lentil physiological maturity, around mid-July in both years, an area of 1.98 m² of each plot (6 inner rows of 2 m long were harvested to exclude edge effects) was carefully hand-harvested at ground level in order to measure the yield and to prevent pod opening and lentil grain loss.

4.2.4. Yield components denomination

For wheat, the consecutive yield components are the number of plants per seed (Plants/Seed) corresponding to the emergence rate, the number of ears per plant (Ears/Plant) corresponding to the fertile tillering rate, the number of grains per ear (Grains/Ear) corresponding to average

spikelet fertility and the individual grain weight (OneGrainWeight) corresponding to the average grain fill achieved. Wheat grain yield per m² can be expressed as the product of the consecutive yield components as follows:

$$\text{Wheat-GrainYield/m}^2 = \text{Seeds/m}^2 \times \text{Plants/Seed} \times \text{Ears/Plant} \times \text{Grains/Ear} \times \text{OneGrainWeight}$$

These yield components allow to calculate “cumulative wheat-yield components” based on surface area such as number of (i) seeds per m² (Seeds/m²), (ii) plants per m² (Plants/m²), (iii) ears per m² (Ears/m²), (iv) grains per m² (Grains/m²) and (v) the yield per m² (Yield/m²).

For lentil, yield components are the number of plants per seed (Plants/Seed) corresponding to the emergence rate, the number of branches per plant (Branches/Plant) corresponding to the branching rate, the number of pods per branch (Pods/Branch) corresponding to flower fertility, the number of grains per pod (Grains/Pod) corresponding to pod fertility and the individual grain weight (OneGrainWeight) corresponding to average grain fill achieved. Lentil grain yield per m² can be expressed as the product of the consecutive yield components as follows:

$$\text{Lentil-GrainYield/m}^2 = \text{Seeds/m}^2 \times \text{Plants/Seed} \times \text{Branches/Plant} \times \text{Pods/Branch} \times \text{Grains/Pod} \times \text{OneGrainWeight}$$

These yield components allow to calculate “cumulative lentil-yield components” based on surface area such as number of (i) seeds per m² (Seeds/m²), (ii) plants per m² (Plants/m²), (iii) branches per m² (Branches/m²), (iv) pods per m² (Pods/m²), (v) grains per m² (Grains/m²) and (vi) the yield per m² (Yield/m²).

4.2.5. Yield components measurement and calculation (Fig. 13)

4.2.5.1. Wheat yield components measurement and calculation

The number of seedlings in 3 rows of 1-meter-long within the 6 inner rows of each plot was counted 1 month after emergence to determine the number of plants per m² (Plants/m²). The number of Seeds/m² corresponds to the number of grains sowed. Then, the Plants/Seed was calculated as the Plants/m² divided by Seeds/m². Other wheat yield components were measured or calculated as follows (Fig. 13). Ears from all of the harvested area were cut and weighed and a subsample of 100 randomly picked up ears was taken and also weighed. The Ears/m² was calculated by dividing the weight of all ears by that of the 100 randomly picked up ears, multiplying by 100 and dividing by the surface area (1.98 m²). Ears from the whole area were

threshed using a research-designed thresher (brand Roland Chateau du Loir, France) to separate grains from straw and debris. Wheat grains were cleaned through an air column and only the heavy sound grain was considered in the results since the small grain fraction was negligible. Heavy sound grains were subsequently oven-dried for 48 hours at 80°C and weighed to determine the Yield/m². A subsample was used to determine the OneGrainWeight. The Grains/m² was estimated by dividing the Yield/m² by the OneGrainWeight. The Grains/Ear was calculated by dividing the Grains/m² by the Ears/m². Finally, the Ears/Plant was calculated by dividing the Ears/m² by the Plants/m².

4.2.5.2. Lentil yield components measurement and calculation

The number of lentil plants per m² (Plants/m²) and Plants/Seed were determined as for wheat (see above). Other lentil yield components were measured or estimated as follows (Fig. 13). Lentils plants from all of the harvested area were threshed using a research-designed thresher (brand Roland Chateau du Loir, France) to separate grains from straw and debris. Lentil grains were composed of two grain fractions (1) the sound grains (heavy grains) and (2) the bruchid-damaged grains (light grains) which were subsequently separated using the same air column as wheat. Grain fractions were oven-dried for 48 hours at 80°C and weighed to determine the dry mass of each fraction (Sound Grain Yield/m² and Bruchid-DamagedGrainYield/m², respectively). Subsamples of each fraction were used to determine respectively the SoundOneGrainWeight and the Bruchid-DamagedOneGrainWeight. The Grains/m² was calculated by dividing each grain fraction mass by its respective OneGrainWeight (corresponding to the SoundGrains/m² and Bruchid-DamagedGrains/m², respectively) and summing the grain fractions (Fig. 13).

In order to calculate yield components, 15 plants of lentil were randomly picked up within the 6 inner rows of each plot but out of the area of 1.98 m² (see above) in both years at the day of harvest (Fig. 13). The number of branches, the number of pods and the number of grains (sound grains and bruchid-damaged grains) were counted to determine the Grains/Pod and the Pods/Branch. Then, the Pods/m² was calculated as the ratio between the Grains/m² and the Grains/Pod and the Branches/m² as the ratio between the Pods/m² and the Pods/Branch. Finally, the Branches/Plant was calculated as the ratio between Branches/m² and the Plants/m² (Fig. 13).

Bruchid damages cause significant and variable losses of lentil grains which makes comparisons of hand-harvested yields inaccurate (Viguier et al. 2018a). Therefore, we

considered the attainable lentil yield ($\text{AttainableYield}/\text{m}^2$) which corresponds, in our situation, to the amount of lentil grain that would be produced in the absence of bruchid damages. We assumed that bruchid larvae damaged developing lentil grains late enough during the crop reproductive phase so that lentil plants could not compensate for the damage, and that lentil yield components (i.e. Grains/m^2 and $\text{SoundOneGrainWeight}$) were similar to those expected in the absence of bruchids. Consequently, the $\text{AttainableYield}/\text{m}^2$ was estimated as the product of the Grains/m^2 and the $\text{SoundOneGrainWeight}$ (Fig. 13). Henceforth, for lentil, the term Yield/m^2 used in the article refers to the $\text{AttainableYield}/\text{m}^2$ and the term OneGrainWeight refers to the $\text{SoundOneGrainWeight}$.

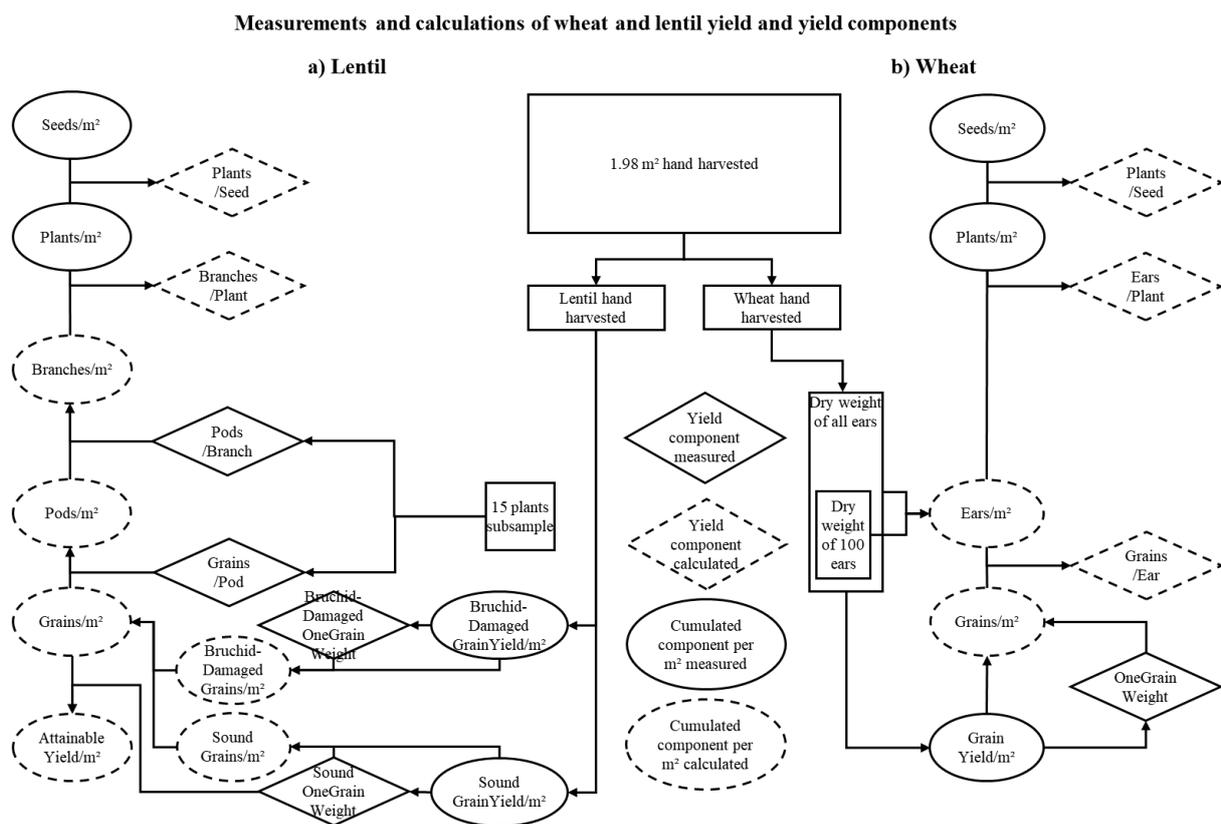


Fig. 13 Diagram explaining how species yield and yield components were measured or calculated for a) lentil and b) wheat. Symbol shapes indicates the type of components (diamond = yield component and oval = cumulative component expressed by unit of surface). Peripheral line type indicates how the component's value was obtained (straight line= measurement and dashed line = calculation). Arrows indicate the sequences of operations.

4.2.6. Calculation of Interspecific interaction indices for yield components

The effect of a species on the companion species in a bi-specific intercrop was evaluated with the interspecific interaction index adapted from Jacquard (1968) by Bedoussac and Justes

(2010). The principle is that the effect of wheat on a yield component of lentil can be evaluated by comparing treatments in which lentil is grown at the same plant density in intercrop and sole crop, and vice-versa for wheat. If the yield component value of lentil is different in intercrop than in sole crop, the difference can be attributed to the effect of wheat, i.e. the interspecific interaction of wheat, on lentil and vice-versa for wheat. Interspecific ratio (IE) for each yield component is the ratio of the yield component measured in intercrop over that measured in sole crop:

$$L-IE_X = \frac{X_{IC}}{X_{SC}} \text{ and } W-IE_X = \frac{X_{IC}}{X_{SC}}$$

with “X” a yield component of lentil for L-IE (Plants/Seed, Branches/Plant, Pods/Branch, Grains/Pod, OneGrainWeight) or of wheat for W-IE (Plants/Seed, Ears/Plant, Grains/Ear, OneGrainWeight) as measured on plots where targeted plant densities are identical in intercrop (IC) and sole crop (SC).

Interspecific ratio per surface unit (IES) for each cumulative yield component is the ratio of the cumulative yield component measured in intercrop over that measured in sole crop:

$$L-IES_X = \frac{X_{IC}}{X_{SC}} \text{ and } W-IES_X = \frac{X_{IC}}{X_{SC}}$$

with “X” a cumulated yield component per surface area of lentil for L-IES (Seeds/m², Plants/m², Branches/m², Pods/m², Grains/m² and Yield/m²) or for wheat for W-IES (Seeds/m², Plants/m², Ears/m², Grains/m² and Yield/m²) as measured on plots where targeted plant densities are identical in intercrop (IC) and sole crop (SC).

Values of IE or IES lower than one indicate that the yield component or the cumulative yield component (per surface unit) is lower in intercrop than that in sole crop due to interspecific competition produced by the companion species, and vice versa. For instance a value of IE or IES equal to 0.8 indicates that the yield component value in intercrop is 20% lower than in sole crop at the same targeted plant density.

4.2.7. Statistics

Shapiro-Wilk and Levene’s tests were used to test the normality of the data and the homoscedasticity of its variance, respectively. Analysis of variance were performed using the AOV procedure of R software v. 2.7.1 (R development Core Team 2007). To avoid comparisons with unbalanced designs on yield components (IES and IE values), only treatment IC100%L:33%W was used for the study of the dependent variables year and cultivar (Table 3)

and only lentil cv. Anicia and Beluga were considered for analysis of the wheat density effect (Table 3). The independent variables: year, cultivar, associated cultivar and bloc were always included in the ANOVA. Cropping system, species and wheat density were tested when necessary (i.e. consistent with the dependent variable). Non-significant effects or interactions were removed from the models. In the results section, we will present only interactions related to interspecific ratio (IE) values since the measurement of each yield component is independent (except for the number of branches per plant which was calculated, cf. 4.2.5.2). Indeed, interspecific ratio per surface unit (IES) values depend partially on previous yield components creating a risk of confusing effects. The assumptions of the ANOVA were checked by visual examination of residuals plotted against predicted values. Tukey's Honest Significant Difference test with $\alpha=0.05$ was used to compare groups's means to allow multiple comparisons in the case of significant effects. Pearson correlations between yield components were calculated using the "cor.test" command of R software. Results are presented as mean \pm 1 standard deviation.

4.3. Results

4.3.1. Lentil and spring wheat yields

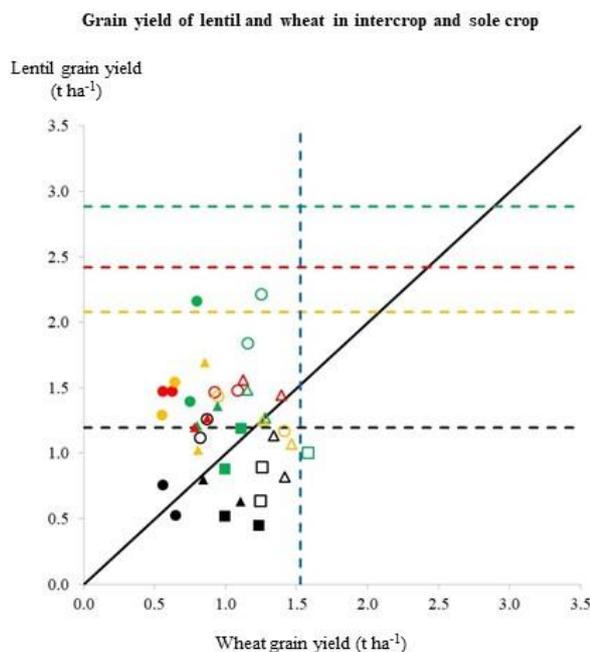


Fig. 14 Lentil grain yield as a function of the wheat grain yield in intercrop and sole crops grain yield. Symbol shape indicates the plant densities (circle = 100%L:17%W, triangle = 100%L:33%W, square = 100%L:50%W). Symbol filling indicates the experimental year (closed = 2015, open = 2016). Symbol color indicates the lentil cultivar (green = Anicia, black = Beluga, red = Rosana, yellow = Flora) while wheat cultivars are not distinguished.

Dashed lines correspond to the mean of SC100% grain yield on both years per cultivar (green = Anicia, black = Beluga, red = Rosana, yellow = Flora, blue = mean of the two wheat cultivars). Straight line corresponds to $Y=X$ and each point corresponds to the mean of three repetitions.

On average of all cultivars over both years, lentil grain yield in intercrop was superior to that of wheat (1.21 ± 0.41 vs 1.02 ± 0.28 t ha⁻¹ respectively; $P<0.001$; Fig. 14). No correlation was found between lentil grain yield and wheat grain yield in intercrop. On average of all situations, total intercrop grain yield was higher than the mean of sole crops (2.23 ± 0.47 vs 1.84 ± 0.38 t ha⁻¹ respectively; $P<0.01$). This was also true when analysing lentil cultivars and years separately except for Beluga in 2015, whose yields were low and not different in intercrop and sole crop (0.64 ± 0.15 t ha⁻¹; Fig. 14). Lentil grain yield was significantly ($P<0.001$) lower in intercrop than in sole crop (2.14 ± 0.75 t ha⁻¹ on average of all cultivars on both years). Wheat intercrop grain yield was also significantly ($P<0.001$) lower than its reference sole crop at a 100% density (1.53 ± 0.10 t ha⁻¹).

Lentil grain yield in intercrop was reduced with increased wheat density (1.41 ± 0.43 , 1.20 ± 0.29 and 0.79 ± 0.27 t ha⁻¹ with 17%, 33% and 50% wheat respectively; $P<0.05$; Fig. 14). Conversely, wheat grain yield in intercrop increased with wheat density (0.85 ± 0.26 , 1.09 ± 0.24 and 1.22 ± 0.19 t ha⁻¹ with 17%, 33% and 50% wheat respectively; $P<0.001$).

Lentil grain yield in sole crop was lower in 2015 than in 2016 (1.82 ± 0.77 vs 2.47 ± 0.67 t ha⁻¹ respectively; $P<0.01$) but no significant difference was found in lentil intercrop grain yield between years (1.14 ± 0.44 vs 1.29 ± 0.36 t ha⁻¹ respectively). Wheat grain yield in sole crop was not significantly different in 2016 than in 2015 but wheat grain yield in intercrop was lower in 2015 than in 2016 (0.82 ± 0.20 vs 1.36 ± 0.16 t ha⁻¹; $P<0.05$).

4.3.2. Effect of the year on lentil and wheat yield components

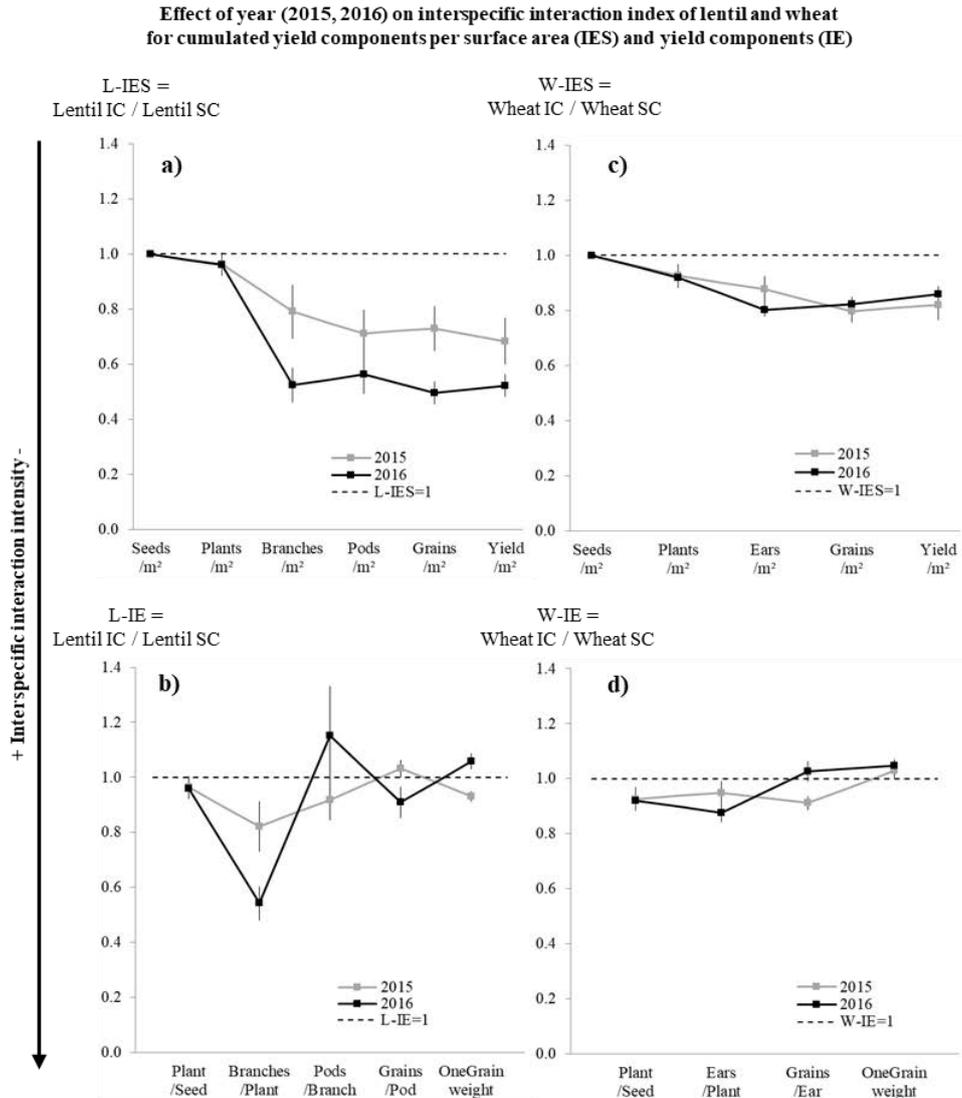


Fig. 15 Effect of the year on interspecific interaction indices of lentil and wheat for yield components in IC100%L:33%W. Mean of four lentil cultivars with both wheat cv. in IC100%L:33%W, (n=8 per year) considering: a) lentil interspecific interaction index for cumulated yield components based on surface area (L-IES), b) lentil interspecific interaction index for yield components (L-IE), c) wheat interspecific interaction index for cumulated yield components based on surface area (W-IES) and d) wheat interspecific interaction index for yield components (W-IE). Vertical bars represent confidence intervals at $\alpha=0.05$.

Interspecific indices involve comparisons at equal sowing densities so that $IES_{Seeds/m^2} = 1$ by definition. The interspecific interaction indices per unit of surface area (IES) for all cumulated components were significantly ($P<0.05$) lower than one for both species on average of both years. This indicates that for lentil, the number of plants, branches, pods, grains and yield per m^2 and for wheat, the number of plants, ears, grains and yield per m^2 were lower in intercrop than in sole crop (Figs. 15a and 15c).

These observations are explained by $L-IE_{Plants/Seed}$, $L-IE_{Branches/Plant}$, $W-IE_{Plants/Seed}$ and $W-IE_{Ears/Plant}$ values lower than one ($P < 0.05$, Figs. 15b and 15d) indicating for both lentil and wheat, a lower number of plants per seed in intercrop than in sole crop, and also for the lentil a lower number of branches per plant and for the wheat a lower number of ears per plant. $L-IE$ and $W-IE$ values for the others yield components were not different from 1 on average of both years.

For lentil, $L-IES_{Plants/m^2}$ of the two years were similar (Fig. 15a) while $L-IES_{Branches/m^2}$, $L-IES_{Pods/m^2}$, $L-IES_{Grains/m^2}$ and then $L-IES_{Yield/m^2}$ were significantly lower in 2016 than in 2015 ($P < 0.01$). For $L-IE$ values, and except for $L-IE_{Plant/Seed}$, all values were alternatively higher and lower, indicating differences in partial compensation in yield components between the two years (Fig. 15b; $P < 0.01$).

For wheat, no effect of year was found on any $W-IES$ and $W-IE$ yield components (Figs. 15c and 15d).

Table 4 Correlation table for the lentil or wheat yield components and yield per m^2 according to the cropping system. a) For intercropped lentil considering the four lentil cultivars over both years in IC100%L:33%W with both wheat cultivars ($n=16$) and for lentil sole crop the four lentil cultivars over both years ($n=8$). b) For intercropped wheat considering the two wheat cultivars over both years in IC100%L:33%W with four lentil cultivars ($n=16$) and in sole crop the two wheat cultivars (33% density) over both years ($n=4$). Correlation values are displayed for the two cropping treatments as IC | SC, and the asterisks *, ** and *** indicate P-values < 0.05 , < 0.01 and < 0.001 respectively.

a) Lentil correlation values (IC SC)						
	Plant/Seed	Branches/Plant	Pods/Branch	Grains/Pod	OneGrainWeight	Yield/ m^2
Plant/Seed	-	-	-	-	-	-
Branches/Plant	-0.37** 0.03	-	-	-	-	-
Pods/Branch	0.19 0.30	-0.47*** -0.61***	-	-	-	-
Grains/Pod	0.12 0.06	0.20 -0.52***	-0.70*** 0.16	-	-	-
OneGrainWeight	0.38** 0.49	-0.44*** 0.04	0.08 0.09	-0.19 0.01	-	-
Yield/ m^2	0.35*** 0.48***	0.41** 0.45*	-0.08 0.10	0.10 -0.05	0.27 0.70***	-

b) Wheat correlation values (IC SC) and their correlation					
	Plant/Seed	Ears/Plant	Grains/Ear	OneGrainWeight	Yield/ m^2
Plant/Seed	-	-	-	-	-
Ears/Plant	-0.22 -0.33	-	-	-	-
Grains/Ear	0.04 -0.20	-0.17 -0.68**	-	-	-
OneGrainWeight	0.10 -0.04	0.55*** 0.83***	-0.17 -0.69**	-	-
Yield/ m^2	0.23 -0.002	0.77*** 0.81***	0.23 -0.42	0.72*** 0.85***	-

Correlation relationships were calculated between the various wheat yield components plus yield per m^2 for each species and for both intercrop and sole crop situations. Significant negative correlations were found between lentil IC successive yield components: (1) number of plant per seed and number of branches per plant, (2) number of branches per plant and the number of pods per branch and (3) number of pods per branch and the number of grains per pod (Table 4a). For lentil SC, also considering IC successive yield components, apart from the number of

branches per plant and the number of pods per branch that were also negatively correlated, the others successive yield components were not related to each others (Table 4a). Yield m^{-2} was positively correlated to the number of plant per seed and the number of branches per plant for both cropping system (Table 4a). The OneGrainWeight was strongly, positively correlated with the Yield m^{-2} for lentil SC and the correlation was weaker but almost significant ($P=0.062$) for lentil SC.

For wheat in intercrop and sole crop, correlations between successive yield components were always negative apart from OneGrainWeight and Yield m^{-2} that were strongly positively correlated (Table 4). Correlations between successive yield components tended to be weaker for wheat IC though and were significant only for OneGrainWeight and Yield m^{-2} (Table 4b). For both wheat in intercrop and sole crop Yield m^{-2} was also positively and strongly correlated to the number of ears per plant (Table 4), indicating that yield was mainly determined by this component.

4.3.3. Effect of wheat density on lentil and wheat yield components

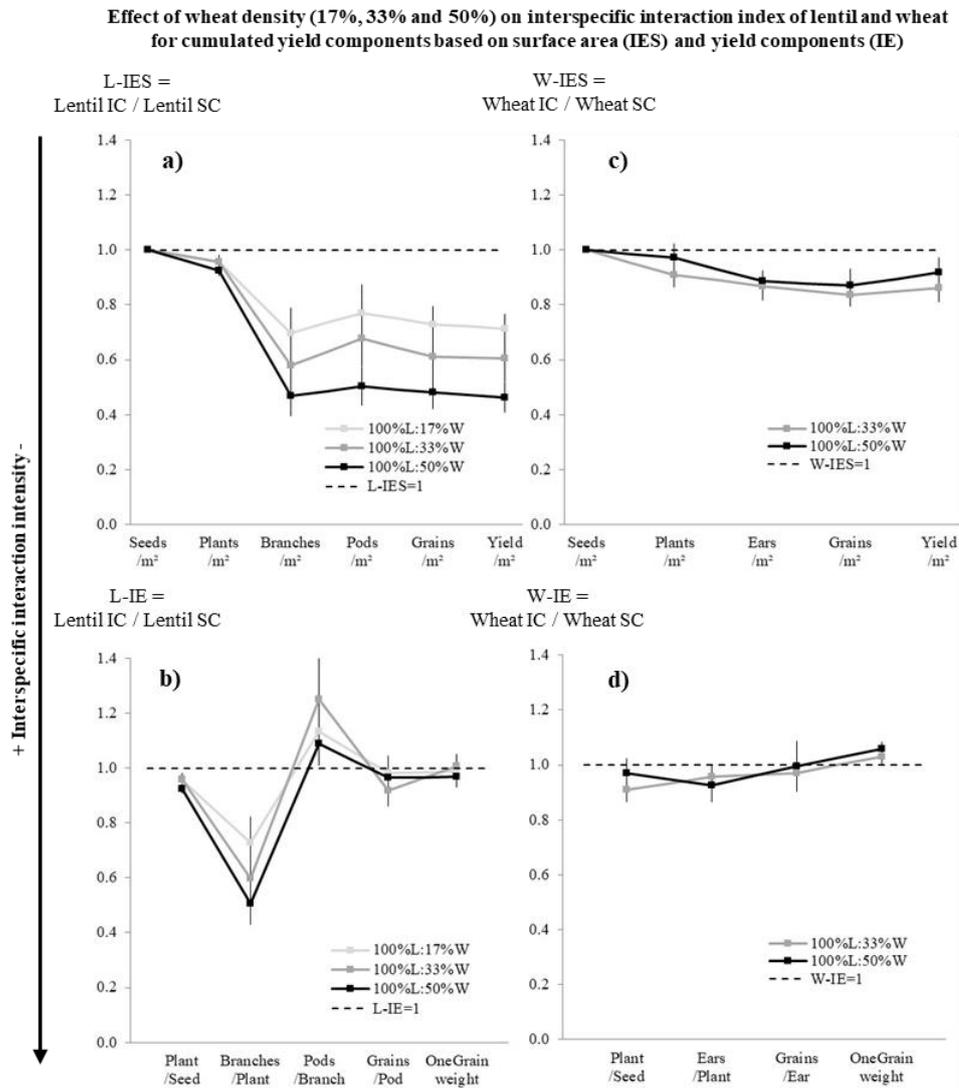


Fig. 16 Effect of wheat density on interspecific interaction indices of lentil and wheat for yield components. Mean of lentil cv. Anicia and Beluga with both wheat cultivars. over both years (n=8 per treatment) considering: a) lentil interspecific interaction index for cumulated yield components based on surface area (L-IES) and b) lentil interspecific interaction index for yield components (L-IE), c) wheat interspecific interaction index for cumulated yield components based on surface area (W-IES) and d) wheat interspecific interaction index for yield components (W-IE). Vertical bars represent confidence intervals at $\alpha=0.05$.

For lentil, L-IES values from L-IES_{Branches/m²} to L-IES_{Yield/m²} were all significantly ($P<0.05$) lower than one; for L-IES_{Plants/m²} they were slightly but significantly lower than one except for IC100%L:17%W (Fig. 16a). Values from L-IES_{Branches/m²} to L-IES_{Yield/m²} in IC100%L:17%W were significantly higher than in IC100%L:50%W, ($P<0.05$) and those of IC100%L:33%W were intermediate between the two other IC treatments. For L-IE no significant difference was

found between IC treatments except for $L-IE_{\text{Branches/Plant}}$ which was higher in IC100%L:17%W than IC100%L:50%W, (Figs. 16a and 16b). However, $L-IE_{\text{Branches/Plant}}$ values were greatly lower than one while $L-IE_{\text{Pods/Branch}}$ values were slightly higher than one ($P < 0.05$; Fig. 16b). Testing modalities specified in Fig. 16 for lentil, no significant interaction between treatment and factors year, cultivar, associated cultivar and bloc for the determination of L-IE values.

For wheat, no difference was found between intercrop treatments on W-IES and W-IE (Figs. 16c and 16d). W-IES values from $W-IES_{\text{Plants/m}^2}$ to $W-IES_{\text{Yield/m}^2}$ were all significantly ($P < 0.05$) lower than one, except for $W-IES_{\text{Plants/m}^2}$ in IC100%L:50%W, ($P = 0.13$; Fig. 16b). In addition, $W-IE_{\text{Ears/Plant}}$ in IC100%L:50%W was the only W-IE value significantly lower than one for both treatments ($P < 0.05$; Fig. 16d), indicating that the number of ears/plant of wheat is the most affected component in intercrop. Testing modalities specified in Fig. 16 for wheat, significant factor interactions were only observed for Year \times Wheat density, Wheat cultivar \times Wheat density and Lentil cultivar \times Wheat density on $W-IE_{\text{Plant/Seed}}$ ($P < 0.01$, $P < 0.001$, $P < 0.05$ respectively).

4.3.4. Effect of lentil cultivar on lentil and wheat yield components

Effect of lentil cultivar (Anicia, Beluga, Rosana, Flora) on interspecific interaction index of lentil and wheat for cumulated yield components based on surface area (IES) and yield components (IE)

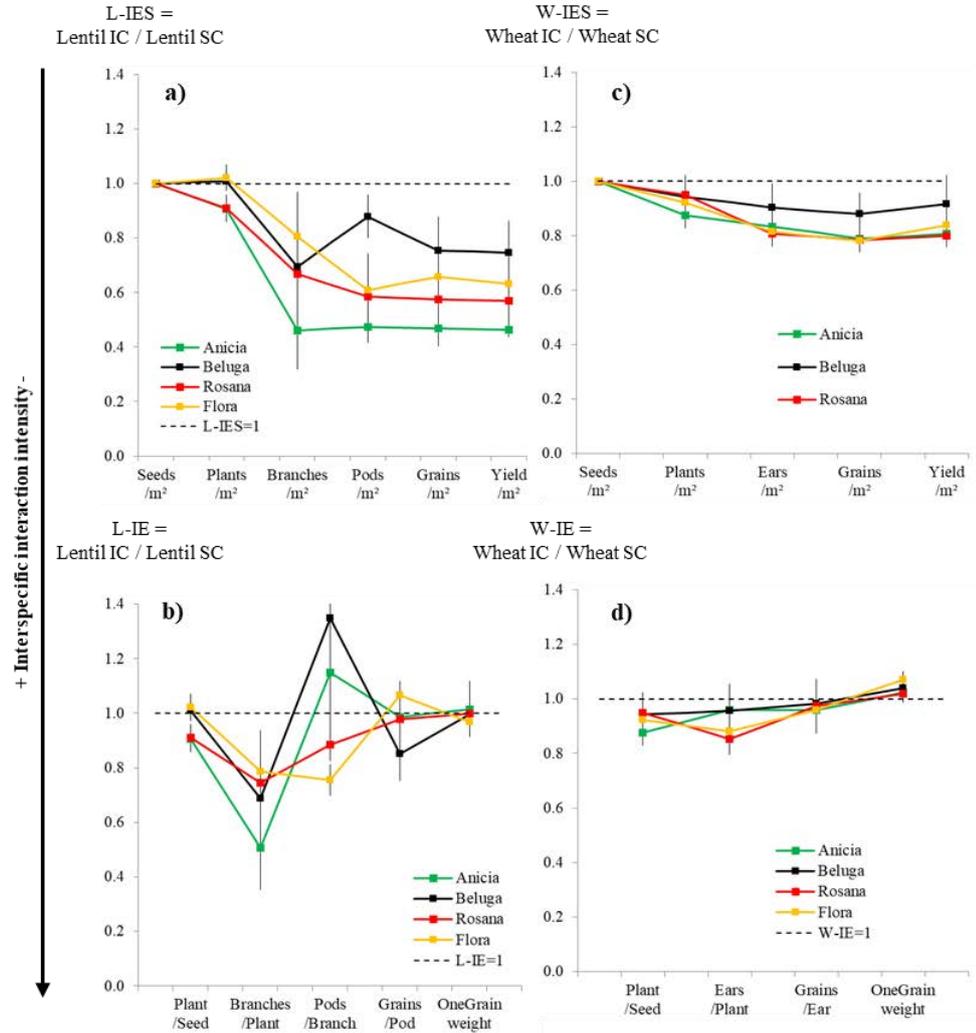


Fig. 17 Effect of lentil cultivar on interspecific interaction indices of lentil and wheat for yield components. Mean of both wheat cultivars. in IC100%L:33%W over both years (n=8 per lentil cultivar) considering: a) lentil interspecific interaction index for cumulated yield components based on surface area (L-IES), b) lentil interspecific interaction index for yield components (L-IE), c) wheat interspecific interaction index for cumulated yield components based on surface area (W-IES) and d) wheat interspecific interaction index for yield components (W-IE). Line and symbol color indicates the cultivar (green = Anicia, black = Beluga, red = Rosana, orange = Flora). Vertical bars represent confidence intervals at $\alpha=0.05$.

For the four lentil cultivars, L-IES values were always lower than one, except for L-IES_{Plants/m²} of cv. Beluga and Flora (Fig. 17a). Lentil cv. Anicia had a lower L-IES_{Branches/m²} than all other cultivars and L-IES_{Branches/m²} of cv. Rosana was also lower than that of cv. Flora (Fig. 17a; $P<0.05$). Lentil cv. Beluga had L-IES_{Pods/m²} higher than all the others lentil cultivars ($P<0.01$;

Fig. 17a), thanks to a $L-IE_{\text{Pods/Branch}}$ almost significantly superior to one ($P=0.07$), and higher than that of cv. Flora ($P<0.05$) and Rosana ($P=0.06$; Fig. 17b). Lentil cv. Beluga was the only cultivar with $L-IE_{\text{Grain/Pod}}$ lower than one ($P<0.05$). Year \times Lentil cultivar interactions were significant for $L-IE_{\text{Plant/Seed}}$, $L-IE_{\text{Pods/Branch}}$, $L-IE_{\text{OneGrainWeight}}$ ($P<0.01$, $P<0.05$ and $P<0.001$ respectively), Year \times Associated wheat cultivar was significant for $L-IE_{\text{Branches/Plant}}$ ($P<0.05$) and Year \times Bloc was significant for $L-IE_{\text{Pods/Branch}}$ ($P<0.001$).

All W-IES values were lower than one, except for $W-IES_{\text{Plants/m}^2}$ and $W-IES_{\text{Yield/m}^2}$ of wheat intercropped with Beluga ($P<0.05^*$; Fig. 17c). W-IES values were globally similar regardless of the associated lentil cultivar. Yet, a significant negative effect of cv. Rosana and Flora was found on $IE_{\text{Ears/Plant}}$ that was lower for wheat intercropped with these two cultivars than with Anicia and Beluga. The interaction Year \times Wheat cultivar was significant for $L-IE_{\text{Plant/Seed}}$ ($P<0.05$) and was the only significant interaction for W-IE.

The effects of wheat cultivar on both lentil and wheat yield components were weak in general (data not shown). However it is worth mentioning that for lentil, L-IES values were lower with Togano than with Valbona ($P<0.05$), due to a stronger reduction of $L-IE_{\text{Branches/Plant}}$ with Togano than with Valbona ($P<0.001$). For wheat, W-IES values were higher with Togano than with Valbona ($P<0.05$), except for $W-IES_{\text{Grains/m}^2}$ which was similar. These results are the consequence of a higher $W-IE_{\text{Plants/Seed}}$ and $W-IE_{\text{OneGrainWeight}}$ for Togano ($P<0.05$) while $W-IE_{\text{Ear/Plant}}$ was similar for the two wheat cultivars.

4.4. Discussion

4.4.1. Global intercrop performances

Intercrops of lentil and spring wheat increase total grain production compared to the mean of reference 100% sole crops (2.23 ± 0.47 vs 1.84 ± 0.38 t ha⁻¹; Fig. 14). This yield advantage has been previously reported in lentil-wheat intercrops in North America (Carr et al. 1995), Bangladesh (Akter et al. 2004), southwestern Germany (Wang et al. 2012), southwestern France (Viguier et al. 2018b) and also on others legume-cereal intercrops, notably in Europe (Hauggaard-Nielsen et al. 2009; Bedoussac et al. 2015). Higher total yields of intercrops are mostly explained by a complementary use of nitrogen sources (soil mineral nitrogen acquisition by the two species and nitrogen from air through N₂ symbiotic fixation by the legume). This situation results in higher total nitrogen amounts accumulated in aboveground biomass of

intercrops versus sole crops and therefore in the total yield advantage (Whitehead et al. 2000; Viguier et al. 2018b).

Both species grain yields were reduced in intercrops as compared to their respective reference sole crops at equal targeted plant density but in different proportions for the two species. The reduction was higher for lentil (-39%) than for wheat (-13%) on average of all treatments and years (Fig. 15) indicating a species difference in sensibility towards interspecific competition. Conversely to Akter et al. (2004 we did not found a negative correlation between lentil and wheat grain yield in intercrops. This result is likely to be caused by the low availability of mineral nitrogen during crop growth in both years that has limited the intercropped wheat biomass and grain yield (yield ranging from 0.55 to 1.59 t ha⁻¹). Consequently, this has certainly narrowed the competitive potential of wheat over lentil so that no relation between the two species grain yields could appear. Interestingly, results from Akter et al. (2004) were obtained from an experimental design with nitrogen fertilizers application at sowing leading to high wheat biomass which increased its competitive ability towards lentil in intercrops. In addition, in a review by Bedoussac et al. (2015) dealing more broadly with legume-cereals intercrops, a negative correlation between cereal and legume yields could be established, likely due to the large range of cereal grain yields in intercrop as a result of many experimental conditions over 10 years and thus a larger range of competition intensity on the legume than in our study.

4.4.2. Early impacts of interspecific interactions on species yield components

The interspecific interaction from wheat to lentil and vice versa began at the very onset of their vegetative phase, i.e. the crop emergence. Indeed, the number of plants per seed of lentil or wheat, corresponding to the emergence rate, was lower in intercrop than in sole crop (minus 4.4% for lentil and 6.1% for wheat). This reduction is low but significant and occurred in a favourable seed-bed preparation in both years. This indicates that in our conditions, the emergence of the companion crop has affected the seed-bed of the other species in a negative way. The lower emergence rates in intercrops could be due to micro-reductions of soil water availability for seeds germination in the intercrop seed-bed due to the higher total amount of seeds sown as compared to sole crops. Indeed, rate of germination for grain legumes and the proportion of germinating seeds decrease with decreasing soil water potential (Kigel et al. 2015). Also, lentil emergence rate can be improved by a process called “seed priming” which consists in water-soaking seeds overnight before sowing (Ali et al. 2009). This process triggers

biochemical changes in seeds, which result in faster, and higher emergence rates thus improves plant stands homogeneity, early vigour and final grain yield (by 29-38%, Ali et al. 2003; Ali et al. 2009). This confirms the importance of adequate amounts of water, at least for lentil seeds, for favourable emergence (Kigel et al. 2015). Allelopathic effects that can affect the processes of germination and emergence of the companion species could also be involved in the emergence reduction in intercrops. The numerous significant interactions on crop emergence observed in our study for both lentil and wheat suggest that crop emergence is a complex trait depending at least on species cultivars, wheat density and environmental conditions. Khalik et al. (2001), in a one-year experiment of alternate rows design for lentil-wheat intercrops and Yagmur and Kaydan (2006) in a line-mixing lentil-barley intercrops reported a comparative significant reduction of plants per m² of both species in intercrops as compared to sole crops. Like us, their experimentation could not provide explanations to this observation so that we suggest this particular question should be addressed in future studies. Nevertheless, this early effect is relatively weak and therefore cannot explain alone the yield difference between intercrops and sole crops for both wheat and lentil.

For lentil, the number of branches per plant was the component most correlated with final grain yield in intercrop (0.41**, Table 4) and therefore the yield component that mainly explains the yield difference between intercrops and sole crops. The important reduction in the number of branches per plant and branches per m² in intercrops (minus 32% and 34% respectively) probably led to the reduction of lentil biomass (minus 41% on average of all treatments and years, data not shown) since the number of branches per plant is positively correlated with biomass (Singh 1977; Aghili et al. 2012). Cereals tend to have a faster above- and belowground development than legumes (Brooker et al. 2014) as demonstrated on barley and pea intercrops (Hauggaard-Nielsen et al. 2001). Therefore, we assume that early competition of wheat for resources acquisition such as light, water and mineral nitrogen has prevented the lentil from accessing to as much resources than in sole crop, leading to a reduced lentil vegetative development. Furthermore, similar values of the number of pods per branch, grains per pod and individual grain weight between lentil intercrop and sole crop indicates that lentil is not able to compensate, by an increase of later yield components, the early decline observed in the number of branches per plant in intercrops.

For wheat, the main yield component that explains the yield difference between intercrops and sole crops is the reduction of the number of ears per plant and ears per m² (minus 9% and 16% respectively on average of both years, Figs. 15c and 15d) indicating an effect of interspecific

competition from lentil on tillering and/or on tiller fertility. This can be also be attributed to the resources acquisition of the other species i.e. in that case, the lentil. A comparable reduction number of ears per m² was observed in intercrops of lentil-barley (Yagmur and Kaydan 2006), faba bean-wheat (Barker and Dennett 2013) and pea-wheat (Bedoussac et al. submitted) suggesting that this feature be typical in legume-cereals intercrops. The physiological effect of the reduction could be either a reduced tillering or the abortion of tillers after tiller initiation or the combination of these two phenomena. Since the number of tillers per plant was not measured, we cannot conclude but we suggest that this particular question could be addressed in future studies.

Similarly to lentil, our results indicate that on average, the intercropped wheat did not compensate the lower number of ears per plant and per surface area by an increased number of grains per ear or by a higher individual grain weight as compared to wheat sole crop. This highlights that (1) the interspecific competition of the lentil in the later development stages is limited since we did not observed lower values in intercrop than in sole crop, but (2) it still has an effect on wheat, preventing the cereal from yield compensation by components determined later during crop growth.

4.4.3. Intensity of interspecific interactions is affected by year

Climatic conditions of 2016 were globally favourable to both species and especially lentil due to the regular rainfall distribution along with softer temperatures during grain filling, a key phase in the determination of final yield (Ali et al. 2009). Conversely, the dry spring in 2015 certainly led to the occurrence of water stress for the lentil during this critical period. Erskine and El Ashkar (1993) and Hamdi (1996) reported that differences in seasonal rainfall could explain up to 80% of the grain yield variability. Consequently, we were expecting a strong effect of year on interspecific interactions and yield components of both species.

For lentil, interspecific interactions were spectacularly more intense in 2016 than in 2015. The main difference occurred on the number of branches per plant which was 46% lower in intercrop than in sole crop in 2016 as compared to a reduction of 18% in 2015 ($P < 0.001$; Figs. 15a and 15b). The difference between years on others IES values remained stable until $L-IES_{Yield/m^2}$ confirming that the number of branches per plant is the main yield component determining the final grain yield.

The number of branches per plant in sole crop was statistically similar in both years but it was lower in intercrop in 2016 than in 2015 ($P < 0.001$). Therefore, the lower $L-IE_{Branches/Plant}$ in 2016

can only be due to a more intense interspecific competition from wheat in 2016 ($P < 0.001$). This was confirmed by a higher number of ears per plant for wheat in intercrop in 2016 than in 2015 ($P < 0.001$). Early competition from wheat for resources acquisition was thus more intense in 2016 and resulted in lower lentil intercrop branching. Nevertheless, all other lentil intercrop yield components (number of pods per branch, grains per pod and OneGrainWeight) were higher in 2016 than in 2015 ($P < 0.05$) suggesting that the compensations mechanisms are related to the year effect (Materne and Siddique 2009; Saxena 2009). Our results indicate that different intensity levels of interspecific interactions can lead to similar lentil grain yields in intercrop confirming that intercrops can improve yield stability of lentil. However, it also suggests that in our conditions of low inputs system, the lentil attainable grain yield in intercrop is limited to a value close to that observed in our trials since favourable environmental conditions would almost always also favour wheat within the intercrop hence strengthening its competitive ability hence its relative impact on lentil.

Regarding the wheat, the intensity of interspecific competition was similar between years but the wheat yield in intercrop was higher in 2016 than in 2015 as a result of higher values for ears per plant and OneGrainWeight ($P < 0.05$). This suggests that even in low inputs system where N is low, N is not the only limiting factor.

4.4.4. Wheat density strongly affects lentil yield in intercrops

The negative correlation between lentil grain yield and cereal plant density as well as the positive correlation between cereal grain yield and cereal plant density in intercrops has been previously observed in lentil-cereal intercrop studies (Khalik et al. 2001, Akter et al. 2004, Yagmur and Kaydan 2006, Viguier et al. 2018b).

Higher wheat density increases the potential of resources acquisition of wheat in intercrop at the expense of that of lentil. Lentil being shorter than wheat, the latter likely causes a shading effect that may be more important with increased wheat plant density limiting the photosynthesis of the legume as shown by Neugschwandtner and Kaul (2014) in oat and pea intercrops. Belowground competition may also explain the yield difference with increased water and mineral nitrogen acquisition by wheat at higher plant densities. Surprisingly, the competition of wheat was already high with only 17% wheat sole crop density (addition of 75 wheat plants per m^2). This could be the result of wheat tillering compensating partially the low cereal density at sowing.

Again, the difference in interspecific competition intensity between wheat densities occurred early, which affected mostly the number of branches per plant and per m² with an increased effect as wheat density increased (Figs. 16a and 16b). Conversely, the other yield components were not affected regardless of the wheat density (Fig. 16b). This confirms that lentil physiology was affected mostly during the vegetative phase whatever the wheat density while the setting up of later yield components remained quite similar to that for sole crops. This last point is consistent with the results of Akter et al. (2004) reporting the absence of effect of wheat density on the individual grain weight.

As for wheat, its density in intercrops had no effect on interspecific competition from lentil (Figs. 16c and 16d). This is due to its ability to compensate low densities by a higher tillering leading, in our limited nitrogen availability conditions, to similar yield potential regardless of the density as confirmed by similar wheat grain yield between SC33%W and SC50%W. This suggests again that cereal grain yield in intercrops is determined more by the environment, than by the interspecific competitions from the lentil.

4.4.5. Sensitivity to interspecific competition varies among genotypes, especially in lentil

Lentil cv. Anicia was the most affected by wheat interspecific competition, Rosana and Flora had intermediate sensitivity and Beluga was the less affected (respectively -54%, -43%, -37%, -25% in grain yield in intercrops compared to sole crop). Again the number of branches per plant and m² was the determining component of the lentil grain yield in intercrop (-49%, -26%, -21% and -31% in number of branches per plant in intercrop compared to sole crop, for Anicia, Rosana, Flora and Beluga respectively). Our results show a diversity in lentil plasticity towards wheat competition in an attempt to compensate interspecific interactions. Conversely to our expectations, lentil cultivars susceptibility or tolerance to wheat competition was not correlated with their precocity (defined as the thermal time needed from sowing to flowering) since Flora and Beluga were respectively the more and the less early.

Interspecific differences in plant traits are a key factor for explaining the positive diversity-productivity relationship in intercropping (Zhu et al. 2015). Therefore we assume that the great variability in interspecific competition response depends on lentil cultivar characteristics, such as height and the ability to produce branches in situations of interspecific competition. We assume that other characteristics such as the leaf area index and the number, orientation and

distribution of leaves also play a significant role in the establishment of competition. For example, light spectrum alterations by plant canopies have been shown to occur quite early, few weeks after emergence, and act as powerful signals for early detection of future competitor neighbour plants and photomorphogenetic responses (Ballaré, 1999).

Wheat was only slightly affected by lentil cultivars as shown by the low range of $W-IES_{Yield/m^2}$ values according to the associated lentil cultivar (0.72-0.94 vs 0.41-0.97 for $L-IES_{Yield/m^2}$ of lentil; Figs. 17a and 17c). None of the lentil cultivars height was higher than wheat in intercrops and we assume that none had a deeper root development than wheat (Saxena 2009). Consequently, wheat always dominated lentil regardless of the lentil cultivar so that their competition intensity on wheat was globally identical. However, it is interesting to notice that wheat yield was similar in sole crop and when intercropped with lentil cv. Beluga. This suggests that some combinations of genotypes may display lower interspecific competition on final grain yield for both species.

Regarding the effect of the wheat cultivar, cv. Togano appeared to be more competitive than cv. Valbona as shown by differences in lentil grain yield when intercropped with a particular wheat cultivar (-46% and -33% in intercrop than in sole crop with Togano and Valbona respectively). Meanwhile, wheat cv. Togano was less sensitive to lentil competition than Valbona, which is consistent with its higher competitive ability towards lentil. The effect of wheat cultivars interspecific competition on lentil is again due to a contrasted effect on lentil branching (-40% and -24% branches per plant in intercrop than in sole crop with cv. Togano and Valbona respectively). This result is quite surprising since both wheat cultivars achieved the same grain yield and since cv. Togano was less precocious than cv. Valbona. These results highlights again that cultivars characteristics must be more deeply analysed in order to better understand genotype-genotype interactions. This will be helpful to determine pairs of cultivars of species which are the most efficient when intercropped.

4.5. Conclusion

Our results confirm that growing lentil in intercrops with spring wheat is an efficient agronomic solution to increase total grain yield in low inputs system although both species had their grain yield reduced in intercrops as compared to the sole crops due to interspecific competitions. Our results highlighted that lentil is more affected than wheat by interspecific interactions and we suggest that spring wheat density should remain as low as possible (ca.15-20%) to avoid a strong reduction of lentil yield. The analysis of grain yield components reveals that lentil-spring

wheat intercrop grain yield is determined early during the vegetative stages because of the reduction of lentil branching and wheat earing. The observed results on yield components also confirmed that year lentil genotype and wheat density affected the intensity. Our study also confirmed that the post-harvest analysis of yield components is a relevant approach to understand the dynamics and mechanisms of dynamic competitive interactions.

Our results suggest that the study of cultivar characteristics and pairs of cultivars suited for intercropping to limit competition could help further improve intercrops performance.

Chapter IV. Synthesis, general discussion and perspectives

The first objective of this thesis was to investigate the performance of lentil-spring wheat intercrops for grain yield production compared to sole crops. The second objective was to understand the main underlying mechanisms that explain the performance as key information supporting (1) knowledge on lentil spring-wheat intercrops that can be used for further research on that specific intercrop or more broadly on legume-cereals intercrops, and (2) the design of efficient intercropping practices for lentil-wheat mixture. This section sums up and discusses the main results of this thesis within the framework of our objectives to conclude on the intercrop option for lentil production. It also provides perspectives that arise from this work.

The yield we refer to in this chapter is, unless otherwise mentioned, the estimated attainable yield, i.e. the sum of the observed actual yield plus the calculated yield loss due to bruchid grain damage.

5.1. Synthesis of main results

5.1.1. Higher nitrogen acquisition and total grain yield of intercrops

In chapter III we showed that the well-known, positive linear relationship between grain yield and nitrogen (N) accumulated in shoots was valid in intercrops for both species, regardless of the year, cultivars and crop arrangement. The intercrop as a whole must thus acquire a high amount of N to achieve high grain yield. Lentil N requirements were ca. twice as much as those of wheat which was consistent with observations of Whitehead et al. (2000) on lentil and wheat sole crops.

In our conditions, lentil-wheat intercrops had a genuine advantage (+16%) for N accumulation in shoots over the mean value of lentil and spring wheat sole crops. This led to a total intercrop grain yield higher than the mean of sole crops (2.2 versus 1.8 t ha⁻¹ respectively; Chapter III and Chapter IV). Intercropping lentil and spring wheat is thus a way to increase yields in organic farming without increasing inputs (e.g. water or organic fertilizers) and the observed mean yield advantage is consistent with that reported in other legume-cereal intercrops (Bedoussac et al. 2015).

The advantage of lentil spring-wheat intercrops was the consequence of an effective complementary use of N pools, as shown in a wide range of legume-cereal intercrops (Naudin et al. 2010). Wheat acquired most of the soil mineral N available in the intercrop (73% of the

total mineral N acquired by the intercrop, Chapter III) so that the lentil had to rely more on biological N₂ fixation to meet its high N requirements as shown by a higher proportion of N derived from air (+19%; Chapter III) for intercrop lentil compared to sole crop lentil.

Total intercrop N acquisition and grain yield were substantially higher than those of wheat sole crop (+57% and +31% respectively). For wheat sole crop, mineral N is the unique source of N and its N acquisition was most certainly limited by the low mineral N available in both years (ca. 25 kg N ha⁻¹). Conversely, intercrops could use N₂ fixation which explains the large difference in N acquisition and grain yield between intercrops and wheat sole crop in our conditions.

Intercrops were less efficient in terms of total N accumulation when compared to lentil sole crop. Indeed, the competition of wheat for resources (light, water, nutrients) certainly coupled with the higher physiological cost of the biological N fixation reduced the lentil intercrop biomass and its total N acquisition compared to lentil sole crop (-43% and -46% respectively). Meanwhile, our results showed that lentil sole crop acquired similar amounts of mineral N to wheat sole crop indicating the ability of this legume to efficiently uptake the soil mineral N while having a good level of nitrogen fixation (70%Ndfa; Chapter III). However, the total grain yield of intercrop was similar to that of lentil sole crop because of the difference in N requirements to produce an equivalent grain yield (Chapter II and III).

5.1.2. Wheat intercrop grain protein concentration is increased

Since producing lentil in intercrops was the targeted agronomic outcome of our work due to its higher market value, we did not want wheat density in our additive or substitutive designs to be too high. Thus, we set wheat intercrop density to the range of 17 - 50% of the farm reference density for wheat sole crop. As stated above, wheat intercrop acquired most of the mineral nitrogen available in intercrops but its grain yield was lower than that of the wheat reference sole crop (Chapter III). This resulted in an average increase of 1 point (12 to 13%) in wheat intercrop grain protein concentration compared to wheat sole crop (Chapter III). This increase was observed in both years (Chapter III) and in almost all situations which suggests that this result is robust in low input conditions (Wang et al. 2013; Bedoussac et al. 2015).

This increase in wheat grain protein concentration may appear marginal but cereal grain with a grain protein concentration superior to 12% is worth a premium price especially in organic farming (Chapter II). Indeed in such a type of agriculture, it is very difficult to obtain high grain

protein concentrations due to the prohibition of chemical fertilizers and the low efficiency of organic fertilizers (Wang et al 2013).

As producing high wheat grain protein concentration in organic agriculture is challenging, it could become a driver of intercropping adoption by farmers, along with that of lentil production. Nevertheless, an economic assessment of intercrops is needed to identify the best compromise, due to the negative correlations between (1) wheat grain yield and grain protein concentration and (2) lentil grain yield and wheat grain yield due to species competition (Chapter III). Note that any legume able to reduce the wheat grain yield altogether with uptaking a limited amount of soil mineral nitrogen will have the same effect so that the increased grain protein content of wheat intercrop is not specific to lentil-wheat intercrops (Wang et al 2013; Bedoussac et al. 2015)

No difference was found on lentil grain protein concentration in intercrops and sole crops even when analysing cultivars separately, which is consistent with the studies of Wang et al. (2013) and Burstin et al. (2011). Nevertheless, our results suggest that interactions exist between lentil grain yield, cultivars and years indicating that the physiological processes of N remobilization into lentil grain are complex (Chapter III).

5.1.3. Interspecific competition began early in vegetative growth

Interspecific (species to species) interactions affected markedly more lentil than wheat (Chapter III, IV), as observed in other lentil-cereal intercropping studies (Akter et al. 2004; Wang et al. 2012; Wang et al. 2013).

The effects of interspecific interactions on both species growth cycles appeared very early, i.e. during crop emergence (Chapter IV). The reduction of the number of plants in intercrops, albeit relatively small but significant, was observed on both years and on both species. This effect is no threat to the agronomic performance of intercrops but would be worth being investigated. Crop implantation is a major factor of the final grain production of lentil (Ali et al. 2003; Ali et al. 2009) and may become more critical in some peculiar conditions or possibly in other legume-cereal intercrops. We proposed a hypothesis of a higher water competition between seeds in intercrops compared to sole crop to explain the reduction in the number of plants in intercrops (Chapter IV) but it needs to be investigated.

For both crops, the major impact of interspecific interactions occurred at an early stage of the vegetative phase (shoot ramification), i.e. branching for lentil and tillering for wheat. However,

for the latter, we were not able to figure out if the true physiological effect of competition was a reduced tillering initiation or tiller abortion or both because the number of ear per tiller was not measured (Chapter IV). The higher competitiveness of wheat in intercrop would be the result of a faster, higher aerial and deeper belowground development than lentil, as we observed in our experiment using mini-rhizotrons and measuring plants height in dynamics (results not shown). Consequently, intercropped wheat would had access to (1) higher light quantity in the absence of any strong shading effect by lentil, and more important, (2) larger depth for soil resources.

On both years, lentil was able to acquire a significant proportion of N from mineral nitrogen uptake in intercrop. This acquisition of resources at the expense of wheat explains the competition that lentil exerted on wheat. Globally both crops were not able to effectively compensate the early reduction in branching for lentil and tillering for wheat leading in yield reduction for both species in intercrops (Chapter III and IV).

5.1.4. Intensity of interactions between plants depends on climatic conditions, densities and cultivars

5.1.4.1. Effect of climatic conditions

Fortunately, the climatic conditions (mean, maximal temperatures and rainfall distribution) of our two experimental years were different enough so we could test partially the response of intercrops to climate variability. The combination of high temperatures and uneven distribution of rainfall in 2015 led presumably to a water stress for both species. In 2016 temperatures were generally milder and rainfall distribution relatively homogeneously distributed throughout the crop cycle.

The main effect of climatic year on intercrops was an increase in interspecific competitions from wheat which were more intense in 2016 than in 2015, i.e. on the year with more favourable climatic conditions (Chapter IV). This was due to an increased wheat intercrop biomass in 2016 that occurred with quite similar mineral nitrogen availability between years. Increased wheat biomass increased wheat competition on lentil because of a higher acquisition of abiotic resources from wheat. Consequently, lentil could not benefit from the favourable climatic conditions to increase its biomass and grain yield as suggested by the higher biomass and grain production of lentil sole crops in 2016 (chapter IV). Our results indicate that different intensity levels of interspecific interactions can lead to similar lentil grain yields in intercrop but this also

suggests that, in low input farm conditions in the same regional area, the lentil grain yield in intercrop could be limited to a range value close to that observed in our trials.

5.1.4.2. Effect of density

Density was an important factor of species productivity in intercrops and thus of their global performance as reported in a review of Brooker et al (2014) regarding different types of intercrops in term of spatial and temporal arrangements and species involved. We observed a negative correlation between lentil grain yield and wheat plant density as well as a positive correlation between wheat grain yield and wheat plant density in intercrops (Akter et al. 2005). These relations were also true for N acquisition suggesting that N nutrition in relation to species density would drive biomass production and then intercrop performance. Higher wheat density increases the potential of resource acquisition of wheat in intercrop at the expense of that of lentil which explains the lower lentil grain yield. Belowground competition could also explain the yield difference with increased water and mineral nitrogen acquisition of wheat at higher plant densities. Competition of wheat was already high with only 17% wheat sole crop density (75 wheat plants per m²). Consequently, in order to maximize lentil grain yield in intercrops while maintaining reduced lentil lodging and increased harvest efficiency, we suggest keeping wheat density as low as possible (ca. 15-20% in practice). Furthermore, the increase wheat grain protein concentration in intercrops was also highest with low wheat density which strengthen the advantages of additive intercrops with low wheat density. Using a wheat density lower than 17% might further reduce wheat competition intensity but obtaining a homogeneous wheat cover would then become very challenging. The main risk of uneven wheat cover is a reduction of the advantage in terms of harvest efficiency of lentil and a reduction in resource competitiveness towards weeds.

In practice, we would not recommend to use substitutive designs (i.e. intercrops in which lentil density is below its 100% reference) for lentil production since wheat competition on lentil was even more intense and resulted in lower lentil yields than in additive designs in most of our treatments. Nevertheless, lentil cultivar Anicia grain yield in intercrop on average of both years was similar in the substitutive design 67%L:33%W to the 100%L:33%W (Chapter III). Thus, the reduction of lentil density in intercrops was not systematically associated with lentil grain yield reduction (Chapter III). Meanwhile, wheat grain yield tended to be negatively correlated with lentil density (Chapter III) but it was not significant probably because intercrop lentil was not able to acquire sufficient amount of resources to significantly reduce wheat grain yield.

5.1.4.3. Effect of genotype

All lentil cultivars were sensitive to the effect of wheat competition in intercrops (Chapter II and III). However, the study of the yield components showed differences in cultivar sensitivity to interspecific interactions and differences in developmental compensations (Chapter IV). Differences in wheat cultivar competitiveness were also observed and the study of interactions suggested that some couples of lentil-wheat cultivars displayed less interspecific interaction intensity and were more complementary. Consequently, this suggests that breeding cultivars suited to intercropping (both lentil and wheat) could be a way to improve the performance of this practice. Finally, we showed that lentil susceptibility to wheat competition did not seem to be related with cultivar precocity (time to flowering; Chapter IV) since both the most and the least early lentil cultivars had the lowest interspecific interaction intensity on grain yield.

5.1.5. Effect of intercrops on lentil agronomic constraints

5.1.5.1. No observed effect of intercrops on bruchid damages

We showed that unfortunately bruchid damage on lentil were not reduced in intercrop with wheat. There was a significant effect of year on bruchid damage rates which was 16% lower on average in 2016. Significant differences were found on bruchid damage rates between cultivars with Anicia having the highest damage, Beluga the lowest, and Rosana and Flora similar intermediate damage. The difference between lentil cultivars was also observed when years were analysed separately. No significant effect of lentil and wheat densities and no significant interactions between factors were found. The yield gap analysis revealed that bruchids consumed on average ca. 25% of the grain yield of lentil in both intercrop and sole crop (Chapter II), which is a high loss of matter and of an even higher significant impact on market value due to the presence of damaged grain within the harvested grain. However, our field experiments were not designed to analyse bruchid damage rates. Consequently, we were only able to provide hypotheses to explain the observed variability associated with bruchid damage rates.

Laserna-Ruiz et al. (2012) found an infestation rate in lentil seeds from 0-70% on a lentil germplasm collection of 571 accessions under field conditions in Spain which is consistent with our high level of seed infestation and variations between cultivars. Differences in bruchid infestation rates among cultivars were also found in broad bean with *Bruchus rufimanus* (Szafirowska 2012), a different species though than those affecting lentils in our conditions.

Variations between cultivars are likely due to differences in host-plant resistance mechanisms (i.e. antibiosis, antixenosis, tolerance and ecological resistance) towards bruchids (Painter 1951; Kogan 1982; Laserna-Ruiz et al. 2012). Szentesi and Jermy (1995) reported that bruchid infestation rates were affected by seed dimensions of the grain legume. In our trial, seeds from lentil cv. Beluga were the smallest and had the lowest thousand grain yield and conversely for lentil cv. Anicia. This might have been an example of an antixenosis (non-preference) defence mechanism towards bruchids from lentil cultivar Beluga (Laserna-Ruiz et al. 2012). Nevertheless, lentil defence mechanisms towards bruchids remain unclear and given the major impact of this beetle on lentil, it seems urgent to start investigations to understand it better (see below, some ideas developed in perspectives).

5.1.5.2. Reduction of late emergence weeds in intercrops

In chapters II, III and IV we mentioned that weeds were handily removed when necessary. One species of weed exerted potentially some pressure in some areas of our field trials in each year (field horsetail, *Equisetum arvense* in 2015 and wild buckwheat, *Fallopia convolvulus* in 2016). These weeds were recurrent in these fields (but we did not have the information before setting up the experiment) and the stale seed-bed before sowing (two uses of spring-tooth harrows) performed each year did not prevent them from emerging. Both weed species were thus removed by hand during the whole crop cycle due to their spatial heterogeneity (mainly in the border of the plot) to make sure they had only a low effect on our experimental results. Nevertheless, the stale seed-bed worked well on early spring weeds since very few emerged concomitantly with lentil and spring-wheat emergence on both years.

The late cycle weeds, (those which appear at the time of lentil flowering or later) were not removed from the plots and were hand harvested at the same time as lentil and wheat. They were separately treated as the crops for dry weight determination. ANOVA of the amount of weeds at harvest was performed to test the effect of the cropping system (SC/IC), species (lentil/wheat) and year.

The amount of weeds at harvest was significantly lower in intercrops than in both sole crop lentil and wheat on average for both years and when years were analysed separately ($P < 0.05$). Globally, weeds biomass production was low representing 1%, 4% and 5% of the harvested biomass of total intercrop, lentil sole crop and wheat sole crop respectively, on average for both years. The effect of intercrops on weed biomass is more spectacular in case of high weed pressure, as confirmed by results of the LEGITIMES project that studies lentil-wheat intercrops

at farmer level in southwestern France. In this project, a mean reduction of 39-47% of weed biomass was observed at harvest in intercrops compared to sole crop (Fig 18). The reduced weed pressure is due to the acquisition of resources of the intercrops (light, water, nutrients) that limit weeds development (Wang et al. 2012).



Fig. 18 Effect of intercrops on weed abundance. On both pictures: left = sole crop lentil and right = intercrop lentil plus wheat. Pictures taken from the LEGITIMES project.

The reduction of weeds can be particularly interesting in the case of lentil production because quality standards on grain sorting and cleaning are particularly high (Yenish et al. 2009). Lentil and weeds that would reach maturity concomitantly would have their grain harvested at the same time thus increasing the amount of undesirable grain to discard in the grain cleaning process. Reduced amounts of weeds can then facilitate sorting for cooperatives, even in the case of intercrops.

Furthermore, in organic farming, agronomic damage caused by important weed biomass can be significant since synthetic herbicides are prohibited. The use of intercrops in rotations can help to reduce the build-up of weed seed bank in fields.

5.1.5.3. Reduction of lentil lodging by intercrop wheat leads to a better lentil harvest ability and a higher intercrop profitability

Our adaptation of the yield gap concept was made to include all grain losses due to mechanical harvest and grain cleaning/sorting (Chapter II). We could therefore detail lentil grain losses along the agronomic production stages from the attainable yield to the marketable yield. As previously stated, total intercrop attainable yield was similar to that of lentil sole crop (Chapter

III). This was also true when considering the actual yield level (i.e. hand harvested yield without correction of the yield reducing factor bruchids). Therefore, in our context of lentil production, using intercrops may at first appear as irrelevant. Moreover, the lentil actual yield decrease in intercrop was not economically offset by the actual yield of spring wheat, given a selling price of lentil ca. four times that of spring wheat (Chapter II). As lentil contributes more to intercrop gross margin, actual gross margin of intercrops based on actual yield were lower than that of lentil sole crop. Akter et al. 2004 studied lentil and wheat in additive intercrops in Bangladesh and concluded that intercrops based on actual yield had a higher monetary advantage than lentil sole crop because total intercrop grain yield was higher than that of lentil sole crop, even though lentil grain yield was also reduced in intercrops compared to lentil sole crop. The economic parameters used in this study were not clear, for instance the selling price of both species was not mentioned. We could guess that it was considered as equal which is unrealistic in our conditions. However, it highlights that a reduction of the price gap between lentil and wheat would strengthen the economic performance of intercrops versus lentil sole crop because wheat would economically compensate more the lentil intercrop actual yield reduction. This would also increase the relative economic performance of wheat sole crops compared to intercrops.

Our analysis of the effect of mechanical harvest on lentil yields showed that using agronomic results based on actual yield in order to evaluate the economic performance of lentil wheat intercrops were not relevant when lentil is mechanically harvested, i.e. systematically in the case of France or Europe. The mean lodging of lentil was strongly reduced in intercrops compared to lentil sole crop (Chapter II) while the mean height of the lowest pod was higher in intercrop than in sole crop (Chapter II). The combination of reduced lodging and higher lowest pod height greatly improved lentil intercrop mechanical harvest efficiency compared to that of sole crop (75% vs. 50%, respectively; Chapter II). After cleaning the harvested yield, we obtained only grain that can be sold, i.e. marketable yield. The marketable yield of intercrop lentil was similar to those of sole crop lentil so when calculating gross margin based on marketable yield, those of intercrops were higher than lentil sole crops because of the addition of wheat grain (Chapter II). An interesting aspect of our economic results was that intercrops had an economic advantage both in the situation of potential lentil crop failure (disease, severe lodging) and in “good” years when lentil yield is high. This indicates that intercrop is a crucial factor for providing more economic stability in organic farming, where a strong inter-annual variability is generally observed for sole crops.

The yield gap analysis revealed that the marketable yield of lentil in intercrop was 42% of its attainable yield and only 28% for lentil sole crop. Comparing both cropping modes led to the conclusion that intercrops are more efficient to convert the lentil attainable yield into marketable grain yield. However, our data suggested that both modes currently produce far below optimum performances (Chapter II). The yield gap analysis illustrated that grain loss at mechanical harvest were an important issue for lentil but clearly highlighted that bruchids were a major reducing factor in our experiments, as is the case for organically farmed lentil in southwestern France.

This extended yield gap is particularly relevant for crops dedicated to human consumption because high quality standards may lead to significant grain discards after harvest. Its rationale could be adapted to a number of crops and help to identify major crop reducing factors with better accuracy with regard to the farmer's reality.

5.2. General conclusion

Intercropping lentil with spring wheat showed important agronomic advantages in conditions of organic farming due to the complementary use of available abiotic resources, as shown for the crucial resource of N. From our observations, the most effective and profitable intercrop mixing ratio to produce a high lentil yield and wheat with high protein concentration is a partial additive design with lentil sown at its reference sole crop density and wheat sown at an optimal low density (15-20% of that in sole crop).

The decisive advantage of intercrops was a reduced lentil lodging which increased harvest efficiency and led to higher gross margins of intercrops based on marketable grain yields than lentil and wheat sole crops. According to our results, intercrops can be considered as a credible option to develop organic lentil production in southwestern France.

Nevertheless, our results showed that both sole crop and intercrop farm lie far below attainable performances in terms of marketable grain production. Bruchids were confirmed to be a major threat to organic lentil production in southwestern France and according to our data, spring wheat did not help to reduce the damage caused by this pest. Further research on bruchids should be implemented in the very near future since farmers are currently helpless.

Finally, our work showed that false conclusions could be drawn when analysing intercrop grain production and gross margins without representing the practical reality. Our extension of the conceptual framework of the yield gap – including a novel definition for mechanically

harvested and marketable grain yields – was designed to mimic farmers' real working conditions and thus greatly increases the application potential of scientific results, as farmers could relate the results directly to their practices. The rationale behind our extension can be used for many types of crops and reducing factors and thus contribute to the development of applied agronomic research.

5.3. Perspectives

5.3.1. Which management of intercrops at the Qualisol cooperative level

Our field results were presented each year to the Qualisol team. Given the positive outcomes of our field trials, especially on lentil lodging reduction, and the enthusiasm of some farmers about potential intercrop adoption, the cooperative Qualisol decided to test intercrops in real farming situations. Feedback from farmers were positive with apparent reduction of lentil lodging and observations of better harvest efficiency. Indeed, when lentil lodges severely, pods open and farmers often observe lentil seedling development at the time of harvest or slightly after. In the case of intercrops, this phenomena was less or not observed. Farmer feedback highlighted that the type of combiner used for harvest could also impact the crop harvestability and that more measurements and R&D investigations were needed to further improve harvest efficiency of intercrop and also sole crop lentil.

Globally, the increase in wheat intercrop grain protein concentration was reported by the cooperative staff in charge of grain quality but with important variations between fields (including low to no increase). This may be due to climat variability between fields that would favour or not spring wheat in intercrops and impact wheat grain protein concentration. Difference in mineral nitrogen availability at sowing may also be an important factor. Our results showed that the intercrop economic performance depended on the addition of wheat with high grain protein content. Thus, intercrops were not as profitable as we reported in the absence of premium price for wheat. In order to avoid this situation, the agronomic advisors increased communication on the necessity of sowing intercrops of lentil-wheat in very low mineral content fields to maximize the practice performance.

The grain sorting of intercrops was a serious issue for the cooperative. Their previous sorting chain was far less efficient for intercrops than lentil sole crops, even with the use of the optical sorter (Chapter II) and, because grains must be sorted rapidly after harvest, the management of the intercrop harvest was a serious problem. Main reasons of the difficulties were inadequate sorting material, and low experience of workers. In 2017, the cooperative invested in a new, higher technology sorting chain that would *in fine* sort intercrops and lentil sole crops at the same speed, and more generally boost the global sorting efficiency of this facility for a variety of crop species and cropping modes. The first feedback on the new sorting chain is encouraging and if sorting efficiency of intercrops increases as planned, it would reduce the sorting costs

difference between intercrops and sole crop lentil and may further increase the profitability of intercrop versus sole crop lentil (Chapter II).

This is an example of how the grain legume lock-in situation as mentioned in the introduction of the thesis (II.3) could be solved. Thanks to the investment of stakeholders (here public subventions, researchers, farmers and cooperatives) the agronomic performance of lentil can be increased and help reduce the reluctance of farmers to grow this crop. Consequently, if efforts from all stakeholders continue to be invested in this crop, its productivity will further increase given the considerable gap between attainable and marketable yields (Chapter II) and lentil production will develop.

5.3.2. Propositions for further research

5.3.2.1. Research on bruchids is needed

The priority for improving lentil cropping systems in southwestern France is to find agro-ecological practices that reduce bruchid damage rates. A better understanding of the insect biology is needed, for instance its behaviour from its grain emergence to mating in the following spring is mostly unknown.

The strong effect of the “year” factor on bruchid damage in our experiments indicates the dependence of bruchid activity to climatic conditions. Moreover, agronomic advisors of Qualisol repeatedly observed important variations in bruchid damages rates between fields in the same year, suggesting a role of the spatial field environment in the behaviour of bruchid. Consequently, we might have to find indicators explaining this variability, by testing the information about (1) field environment and (2) cropping practices, and their interaction. This basic knowledge is needed to design relevant agro-ecological strategies to, if possible, control bruchid damage. Several indicators can be tested, such as minimal distance to the semi-natural habitats, to lentil grain silo, and/or to the nearest lentil field, and also agronomic indicators such as number of years since the last lentil cultivation, use of tillage or rotation length. In case correlations can be established with the bruchid damage rate, such indicators could be used by 1) researchers to look for causal relationships; 2) advisors and farmers to adapt their practices towards agroecological managements more efficient in bruchids control.

Furthermore, we observed parasitoid wasps both years in our fields and coming out of bruchid-damaged grains. We were not able to formally identify these insects but they were most likely of the triaspis species (Hymenoptera: Braconidae). These parasitoids can help to partially

suppress bruchid populations but they cannot control bruchid damage since they lay eggs in developing bruchid larvae so that the grain is already damaged. However, they may help reduce bruchid populations (Laserna-Ruiz et al. 2012). A better understanding of the natural predators or insects that regulate adult bruchid populations is then desirable to be able to adapt cropping practices to maximize biological bruchid regulation.

Finally, the breeding of lentil cultivars with host-plant resistance towards bruchid may also be an option. Laserna-Ruiz et al. (2012) identified potential sources of resistance to bruchid in eleven cultivars of *L. culinaris* and in four cultivars of the wild species of lentil *L. orientalis* in Spain. Their study suggested that if efforts are invested in lentil breeding for bruchid resistance, they should make emphasis on the development of insect-resistance cultivars displaying more than one type of resistance to obtain durable resistance.

5.3.2.2. Further knowledge on lentil eco-physiology in relation to cultivar diversity is also needed

Chapter IV of this thesis showed different sensitivities in lentil cultivars to the competition with wheat, in interaction with year, highlighting potential Genotype x Environment x Management interactions. Our results also suggest that some lentil-wheat cultivar couples show lower intensities of interspecific competition. Consequently, we might want to study in more detail some eco-physiological traits of lentil and spring wheat cultivars in response to intercropping and the environment, to understand which trait expressions are linked with lower competition and/or greater complementarity or facilitation.

For instance, we suggest eco-physiological traits in relation to light use such as leaf width or orientation and other plant / canopy traits that may influence the light use efficiency in intercrops such as plant height or soil covering. Indeed, in this thesis, we found that nitrogen resource was a main factor explaining the performance of intercrops but competition for light or water are also key determinant of intercrop performances (Brooker et al. 2014), especially for lentil whose height is low compared to cereals and which is sensitive to water stress. Ideally, measurements of eco-physiological traits of interest would be analysed in different environments to improve the robustness of the results.

Finally modelling could be a way to help analyse plant interactions in intercrop for having a dynamic view of the interspecific interactions. A significant effort is being developed by European teams (for example in the ReMIX H2020 program) to design models that reasonably

catch the complex behaviour of intercrops, by either improving existing plant-soil crop models (such as STICS-intercrop; Launay et al. 2009) or implementing other formalisms from functional-structural plant models and individual-based models. The data obtained in this work could then be later completed with simulation studies made possible by the modelling advances.

Bibliography

- Aghili P, Imani A A, Shahbazi H, Alaei Y (2012) Study of correlation and relationships between seed yield and yield components in lentil (*Lens culinaris* Medik). *Ann of Biol Res* 3:5042–5045
- Akter N, Alim M A, Islam M M, Naher Z, Rahman M, Hossain I (2004) Evaluation of mixed and intercropping of lentil and wheat. *J Agron* 3:48–51. doi:10.3923/ja.2004.48.51
- Ali O, Sarkar A, Rahman M Gahoonia T S (2003) Lentil cultivation under relay cropping in Bangladesh. *Krishi Katha* 3:12–16
- Ali M, Singh K K, Pramanik S C, Ali M O (2009) Cropping systems and production agronomy. In: Erskine W, Muehlbauer F J, Sarker A, Sharma B (eds) *The Lentil Botany, Production and Uses*. Cabi 213–228
- Andow DA (1991) Vegetational diversity and arthropod population response. *Annu Rev Entomol* 36:551–586
- Andrews M and McKenzie B A (2007) Adaptation and Ecology In: Yadav S S, McNeil D L, Stevenson P C (eds) *Lentil: an Ancient Crop for Modern Times*. Springer Heidelberg 23–32
- Angus J F, Kirkegaard J A, Hunt J R, Ryan M H, Ohlander L, Peoples M B (2015) Break crops and rotations for wheat. *Crop Pasture Sci* 66:523–552. doi:10.1071/CP14252
- Arthur W B (1994) *Increasing Returns and Path Dependence in the Economy*. University of Michigan Press
- Ashraf M, Bokhari M H, Chishti S N (1992) Variation in osmotic adjustment of accessions of lentil (*Lens culinaris* Medic) in response to drought stress. *Acta Botanica Neerlandica* 41:51–62
- Baird J M, Shirtliffe S J, Walley F L (2009) Optimal seeding rate for organic production of lentil in the northern Great Plains. *Can J Plant Sci* 89:1089–1097. doi:10.4141/CJPS08226
- Ball R A, Hanlan T G, Vandenberg A (2006) Stem and canopy attributes that affect lodging resistance in lentil. *Rev Can Phytotech* 86:71–81. doi:10.4141/P05-037
- Ballaré C L (1999) Keeping up with the neighbours: phytochrome sensing and other signalling mechanisms. *TIPS* 4:97–102. doi: 10.1016/S1360-1385(99)01383-7
- Barbosa P, Hines J, Kaplan I, Martinson H, Szczepaniec A, Szendrei Z (2009) Associational resistance and associational susceptibility: having right or wrong neighbors. *Annu Rev Ecol Evol Syst* 40:1–20. doi:10.1146/annurev.ecolsys.110308.120242
- Barker S and Dennett M D (2013) Effect of density, cultivar and irrigation on spring sown monocrops and intercrops of wheat (*Triticum aestivum* L.) and faba beans (*Vicia faba* L.). *Europ J Agronomy* 51:108–116
- Bedoussac L, Justes E (2010) The efficiency of a durum wheat-winter pea intercrop to improve yield and wheat grain protein concentration depends on N availability during early growth. *Plant Soil* 330:19–35. doi:10.1007/s11104-009-0082-2

- Bedoussac L, Hauggaard-nielsen H, Naudin C, Jensen E S (2014) Ecological principles underlying the increase of productivity achieved by cereal-grain legume intercrops in organic farming. A review. *Agron Sust Dev* 35:911–935. doi:10.1007/s13593-014-0277-7
- Bedoussac L, Journet E P, Justes E (2018) A method based on yield components analysis to assess intraspecific and interspecific plant-plant interactions in intercrops. Submitted
- Brooker R W, Bennett A E, Cong W, Daniell T J, George T S, Hallett P D, Hawes C , Iannetta P P, Jones H G, Karley A J, Li L , McKenzie B M, Pakeman R J, Paterson E , Schöb C , Shen J , Squire G , Watson C A, Zhang C, Zhang F, Zhang J, White P J (2015) Improving intercropping: a synthesis of research in agronomy, plant physiology and ecology. *New Phytol* 206:107–117. doi:10.1111/nph.13132
- Büchi L, Gebhard C L, Liebisch F, Sinaj S, Ramseier H, Charles R (2015) Accumulation of biologically fixed nitrogen by legumes cultivated as cover crops in Switzerland. *Plant Soil* 163:175–393. doi:10.1007/s11104-015-2476-7
- Burstin J, Gallardo K, Mir R R, Varshney R K, Duc G (2011) Improving protein content and nutrition quality. In: Pratap A and Kumar J (eds) *Biology and breeding of food legumes*, 314–328. doi:10.1079/9781845937669.0314
- Carr P M, Gardner J C, Schatz B G, Zwinger S W, Guldan S J (1995) Grain yield and weed biomass of a wheat-lentil intercrop. *Agron J* 87:574–579. doi:10.2134/agronj1995.00021962008700030030x
- Cellier P (2017) II. Cycle biogéochimique de l'azote. In: Colomb B (ed) *Guide de la fertilisation raisonnée*. Editions France Agricole 105–119
- Chardigny J, Walrand S (2016) How might oil seeds help meet the protein challenge? *EDP Sci* 23–4. doi:10.1051/ocl/2016019
- Connolly J, Wayne P, Murray R (1990) Time course of plant-plant interactions in experimental mixtures of annuals-Density, frequency, and nutrient effects. *Oecologia* 82:513–526
- Consultative Group on International Agriculture Research (CGIAR) (2014) Lentil. <http://www.cgiar.org/our-research/crop-factsheets/lentil/>. Accessed 15 Mar 2018
- Crutzen P J (2006) The “Anthropocene”. In: Ehlers E, Krafft T (eds) *Earth System Science in the Anthropocene*. Springer, Berlin, Heidelberg 13–18. doi:10.1007/3-540-26590-2_3
- Delobel B (2005) Les plantes hôtes des bruches (Coleoptera Bruchidae) : données nouvelles et corrections. *Bull Mens Soc Linn Lyon* 74:277–291
- De Wit C T, Van Den Bergh J P (1965) Competition between herbage plants. *Neth J agric Sci* 212:221–13
- Diekmann J and Al-Saleh Y (2009) Mechanization. In: Erskine W, Muehlbauer F J, Sarker A, Sharma B (eds) *The Lentil Botany, Production and Uses*. Cabi 248–261
- Duc G, Agrama H, Bao S, Berger J, Bourion V, De Ron A M, Gowda C L L, Mikic A, Millot D, Singh K B, Tullu A, Vandenberg A, Vaz Patto M C, Warkentin T D Zong X (2015) Breeding Annual Grain Legumes for Sustainable

Agriculture: New Methods to Approach Complex Traits and Target New Cultivar Ideotypes, *Critical Reviews in Plant Sciences*, 34:381–411. doi:10.1080/07352689.2014.898469

Duru M, Justes E, Falconnier G, Journet E P, Triboulet P, Magrini M B (2017) Analyse du concept de santé globale pour accompagner les transitions agricoles et alimentaires: application au cas des légumineuses. *Agron Env Society* 7:83–95

Erisman J W, Sutton M A, Galloway J, Klimont Z, Winiwarter W (2008) How a century of ammonia synthesis changed the world. *Nature Geoscience* 1:636–639. doi:10.1038/ngeo325

Erskine W, Goodrich W J (1988) Lodging in lentil and its relationship with other characters. *Can J Plant Sci* 68:929–934

Erskine W, Rihawe S, Capper B S (1990) Variation in lentil straw quality. *Animal Feed Science and Technology* 28:61–69

Erskine W, Diekmann J, Jegatheeswaran P, Salkini A, Saxena M C, Ghanaim A, Ashkar F E L (1991) Evaluation of lentil harvest systems for different sowing methods and cultivars in Syria. *J Agric Sci* 117:333–338. doi:10.1017/S0021859600067071

Erskine W, El Ashkar F (1993) Rainfall and temperature effects on lentil (*Lens culinaris*) seed yield in the Mediterranean environment. *J Agric Sci* 121:347-354

Erskine W, Muehlbauer F J, Sarker A, Sharma B (2009) Introduction. In: Erskine W, Muehlbauer F J, Sarker A, Sharma B (eds) *The Lentil Botany, Production and Uses*. Cabi 1–4

Erskine W, Sarker A, Kumar S (2011) Crops that feed the world 3. Investing in lentil improvement toward a food secure world. *Food Sec* 3:127–139. doi:10.1007/s12571-011-0124-5

Erskine W, Sarker A, Kumar S (2016) Lentil: Breeding. *Encyclopedia of Food Grains* (2nd ed.). Elsevier Ltd. doi:10.1016/B978-0-12-394437-5.00210-2

European Parliament (2018) Texts Adopted: A European strategy for the promotion of protein crop. 1–11. <http://www.europarl.europa.eu/sides/getDoc.do?pubRef=-//EP//TEXT+TA+P8-TA-20180095+0+DOC+XML+V0//EN&language=EN>

Evans L T (1994) Crop Evolution, Adaptation, and Yield. *Camb Univ Press* 30:370. doi:10.1017/S0014479700024509

FAOSTAT (2013) Statistical database of the United Nations Food and Agriculture Organization, Rome. <http://www.fao.org/faostat/en/#data/>. Accessed 15 Mar 2018

FAOSTATS (2014) Data on crops. <http://www.fao.org/faostat/fr/#data/QC>. Accessed 05 December 2017

FAOSTAT (2018) Statistical database of the United Nations Food and Agriculture Organization, Rome. <http://www.fao.org/faostat/en/#data/>. Accessed 01 Jun 2018

- Faris M A E, Tahriri H R, Issa A Y (2012) Role of lentils (*Lens culinaris L.*) in human health and nutrition: a review. *Mediterr J Nutr Metab* 6:3–16. doi: 10.1007/s12349-012-0109-8
- Fletcher L A, Kirkegaard A J, Peoples B M, Robertson J M, Whish J, Swan D A (2016) Prospects to utilise intercrops and crop variety mixtures in mechanised, rain-fed, temperate cropping systems. *Crop Pasture Sci* 67:1252–1267. doi:10.1071/CP16211
- Gate P and Gouache D (2010) Les causes du plafonnement du rendement du blé en France : d’abord une origine climatique. Académie d’Agriculture de France. Séance du 5 mai
- German R N, Thompson C E, Benton T G (2016) Relationships among multiple aspects of agriculture’s environmental impact and productivity: a meta-analysis to guide sustainable agriculture. *Biol Rev* 82:716–738. doi:10.1111/brv.12251
- Ghanem M E, Marrou H, Soltani A, Kumar S, Sinclair T R (2015) Lentil variation in phenology and yield evaluated with a model. *Agro J* 107:1967–1977. doi:10.2134/agronj15.0061
- Hamdi A, Erskine W, Gates P (1991) Relationships among economic characters in lentil. *Euphytica* 109:116–57
- Hauggaard-Nielsen H, Ambus P, Jensen E S (2001) Interspecific competition, N-use and interference with weeds in pea-barley intercropping. *Field Crops Res* 101:109–70
- Hauggaard-Nielsen H, Gooding M, Ambus P, Corre-Hellou G, Crozat Y, Dahlmann C, Dibet A, von Fragstein P, Pristeri A, Monti M, Jensen E S (2009) Pea-barley intercropping for efficient symbiotic N₂-fixation, soil N acquisition and use of other nutrients in European organic cropping systems. *Field Crops Res* 113:64–71. doi:10.1016/j.fcr.2009.04.009
- Ibrahim M, Erskine W, Hanti G, Fares A (1993) Lodging in lentil as affected by plant population, soil moisture and genotype. *Exp Agric* 29:201–206. doi:10.1017/S0014479700020640
- Intergovernmental Panel on Climate Change (IPCC) (2014) Climate Change Synthesis Report Summary for Policymakers. 1–32
- Ittersum M K Van, Cassman K G, Grassini P, Wolf J, Tittonell P, Hochman Z (2013) Yield gap analysis with local to global relevance – A review. *F Crop Res* 143:4–17. doi:10.1016/j.fcr.2012.09.009
- Iverson A L, Marin L E, Ennis K K, Gonthier D J, Connor-Barrie B T, Remfert J L, Cardinale B J, Perfecto I (2014) Do polycultures promote win-wins or trade-offs in agricultural ecosystem services? A meta-analysis. *J of Applied Eco* 51:1593–1602. doi:10.1111/1365-2664.12334
- Jacquard P (1968) Manifestation et nature des relations sociales chez les végétaux supérieurs. *Oecologia Plantarum* 3:137–168
- Jensen L S, Schjørring J K, Van Der Hoek K W, Poulsen H D, Zevenbergen J F, Pallière C, Lammel J, Brentrup F, Jongbloed A W, Willems J, Grinsven H (2011) “Benefits of Nitrogen for Food, Fibre and Industrial Production.” European Nitrogen Assessment. Cambridge University Press, 32–61

- Jeuffroy M H, Baranger E, Carrouée B, de Chezelles E, Gosme M, Hénault C, Schneider A, Cellier P (2013) Nitrous oxide emissions from crop rotations including wheat, oilseed rape and dry peas. *Biogeosciences* 10:1787–1797. doi:10.5194/bg-10-1787-2013
- Karel A K, Lakhani D A, Ndunguru B J (1982) Intercropping of maize and cowpeas: effect of plant population on insect pests and yield. *Proceedings of the Second International Symposium on Semi-Arid Areas* 102–109
- Khalik A, Bismillah Khan M, Farrukh Saleem M, Zamir S I (2001) Lentil yield as influenced by density of wheat intercropping. *J of Res* 2:159–162
- Kigel J, Rosental L, Fait A (2015) Seed physiology and germination of grain legumes. In: De Ron Antonio M (ed) *Grain legumes*. Springer 327–363
- Kinane J, Lyngkjaer M F (2003) Effect of barley-legume intercrop on disease frequency in an organic farming system. *Plant Prot Sci* 38:227–231
- Kropff M J, Weaver S E, Smits M A (1992) Use of ecophysiological models for crop-weed interference: relations amongst weed density, relative time of weed emergence, relative leaf area, and yield loss. *Weed Science* 40:296–301
- Lascialfari and Magrini (2016) Towards more sustainable diets: insights from firms' innovation dynamics on new legumes-based food products. *Journées de recherches en sciences sociales*. Dec 2016, Paris, France
- Laserna-Ruiz I, de Los Mozos Pascual M, Santana Méridas O, Sánchez Vioque R, Rodríguez Conde M F (2012) Screening and selection of lentil (*Lens miller*) germplasm resistant to seed bruchids (*Bruchus* spp.). *Euphytica* 188:153–162. doi:10.1007/s10681-012-0752-7
- Laurance W F, Sayer J, Cassman K G (2014) Agricultural expansion and its impacts on tropical nature. *Trends Ecol Evol* 29:107–116. doi:10.1016/j.tree.2013.12.001
- Leroi B, Alzouma I, Huignard J (1990) The influence of intercropping millet (*Pennisetum typhoides* Burm.) with cowpeas (*Vigna unguiculata* (Walp.)) on the egg-laying and development of *Bruchidius atrolinearus* (Pit) (Coleoptera: Bruchidae). *Agric Ecol Environ* 31:39–48
- Letourneau D K, Armbrrecht I, Rivera B S, Lerma J M, Carmona E J, Daza M C, Escobar S, Galindo V, Gutierrez C, Lopez S D, Mejia J L, Rangel A M A, Rangel J H, Rivera L, Saavedra C A, Torres A M, Trujillo A R (2011) Does plant diversity benefit agroecosystems? A synthetic review *Ecol Appl* 21:9–21. doi:10.1890/09-2026.1
- Magrini M B, Anton M, Cholez C, Corre-Hellou G, Duc G, Jeuffroy M H, Meynard J M, Pelzer E, Voisin A S, Walrand S (2016) Why are grain-legumes rarely present in cropping systems despite their environmental and nutritional benefits? Analyzing lock-in in the French agrifood system. *Ecological Economics* 152:162–126. doi:10.1016/j.ecolecon.2016.03.024
- Materne M and Siddique K H M (2009) Agroecology and crop adaptation. In: Erskine W, Muehlbauer F J, Sarker A, Sharma B (eds) *The Lentil Botany, Production and Uses*. Cabi 47-63

- McKenzie B A, Hill G D (2004) Water use in grain legumes. In: Proceedings of the 5th European Conference on Grain Legumes/ 2nd International Conference on Legume Genomics and Genetics, AEP – l'Association Européenne de Recherche sur les Protéagineuse, Paris, France, 61:62
- McKenzie, B A, Andrews M, Hill G D (2007) Nutrient and irrigation management. In: Yadav S S, McNeil D L, Stevenson P C (eds) Lentil: an Ancient Crop for Modern Times. Springer Heidelberg 145–158
- McNeil D L and Materne M A (2007) Rhizobium management and nitrogen fixation. In: Yadav S S, McNeil D L, Stevenson P C (eds) Lentil: an Ancient Crop for Modern Times. Springer Heidelberg 127–143
- Meynard JM, Messéan A, Charlier A, Charrier F, Fares M, Le Bail M, Magrini M B, Savini I (2013) Freins et leviers à la diversification des cultures : étude au niveau des exploitations agricoles et des filières. INRA. doi:10.1051/ocl/2013007
- Monti L M, Grillo S (1983) Legume seed improvement for protein content and quality. Qual Plant Foods Hum Nutr 253:266–32
- Monti M, Pellicanò A, Santonoceto C, Preiti G, Pristeri A (2016) Yield components and nitrogen use in cereal-pea intercrops in Mediterranean environment. Field Crops Res 196:379–388. doi:10.1016/j.fcr.2016.07.0170378-4290
- Muehlbauer F J, Summerfield R J, Kaiser W J, Clement S L, Boerboom C M, Welsh-Maddux M M, Short R W (1998) Principles and Practice of Lentil Production. United States Department of Agriculture (USDA) Agricultural Research Service (ARS) 141:20
- Müller B, and Kreuer D (2016) Ecologists should care about insurance, too. Trends in Ecology & Evolution 31:1–2. doi.org/10.1016/j.tree.2015.10.006
- Naudin C, Aveline A, Corre-Hellou G, Dibet A, Jeuffroy M H, Crozat Y (2009) Agronomic analysis of the performance of spring and winter cereal-legume intercrops in organic farming. J Agric Sci Technol 3:17–28
- Naudin C, Corre-Hellou G, Pineau S, Crozat Y, Jeuffroy M H (2010) The effect of various dynamics of N availability on winter pea-wheat intercrops: crop growth, N partitioning and symbiotic N₂ fixation. Field Crops Res 119:2–11. doi:10.1016/j.fcr.2010.06.002
- Neugschwandtner R W and Kaul H P (2014) Sowing ratio and N fertilization affect yield and yield components of oat and pea in intercrops. Field Crops Res 155:159–163. doi:10.1016/j.fcr.2013.09.010
- Nguyen M L (2018) Nitrogen Fixation by Pulse Crops and the Use of Nitrogen Isotopic Techniques to Measure the Fixation Capacity. In: Rattan L, Stewart B A (eds) Soil nitrogen uses and environmental impacts 21–37
- Olubayo F M, Port G R (1997) The efficacy of harvest time modification and intercropping as methods of reducing the field infestation of cowpeas by storage bruchids in Kenya. J Stored Product Res 4:271–276
- Peoples M B, Brockwell J, Herridge D F, Rochester I J, Alves B J R, Urquiaga S, Boddey R M, Dakora F D, Bhattarai S, Maskey S L, Sampet C, Rerkasem B, Khan D F, Hauggaard-Nielsen H, Jensen E S (2009) The contributions of nitrogen fixing crop legumes to the productivity of agricultural systems. Symbiosis 48:1–17

- Plumecocq G, Debril T, Duru M, Magrini M H, Sarthou J P, Therond O (2018) The plurality of values in sustainable agriculture models: diverse lock-in and coevolution patterns. *Ecology and society* 1:23. doi:10.5751/ES-09881-230121
- Polak R, Phillips E M, Campbell A (2015) Legumes: Health benefits and culinary approaches to increase intake. *Cli Diabetes J* 198:205–33. doi:10.2337/diaclin.33.4.198
- Preissel S, Reckling M, Schläfke N, Zander P (2015) Magnitude and farm-economic value of grain legume pre-crop benefits in Europe: a review. *Field Crop Res* 175:64–79. doi:10.1016/j.fcr.2015.01.012
- Quinn M A (2009) Biological Nitrogen Fixation and Soil Health Improvement. In: Erskine W, Muehlbauer F J, Sarker A, Sharma B (eds) *The Lentil Botany, Production and Uses*. Cabi 229–247
- Rahman M M, Sarker A, Kumar S, Ali A, Yadav N K, Rahman M L (2009) In: Erskine W, Muehlbauer F J, Sarker A, Sharma B (eds) *The Lentil Botany, Production and Uses*. Cabi 121–136
- Raseduzzaman M D and Jensen E S (2017) Does intercropping enhance yield stability in arable crop production? A meta-analysis. *Europ J of Agron* 91:25–33. doi:10.1016/j.eja.2017.09.00
- Rauber R, Schmidtke K, Kimpel-Freund H (2001) The performance of pea (*Pisum sativum* L.) and its role in determining yield advantages in mixed stands of pea and oat (*Avena sativa* L.). *J. Agron. Crop Sci* 187:137–144. doi: 10.1046/j.1439-037X.2001.00508.x
- Reckling M, Hecker J M, Bergkvist G, Watson C A, Zander P, Schläfke N, Stoddard F L, Eory V, Topp C F E, Maire J, Bachinger J (2016) A cropping system assessment framework—Evaluating effects of introducing legumes into crop rotations. *Europ J Agron* 76:186–197. doi:10.1016/j.eja.2015.11.005
- Reda A (2015) Lentil (*Lens Culinaris Medikus*) Current status and future prospect of production in Ethiopia. *Adv Plant Agric Res* 2:1–9. doi:10.15406/apar.2015.02.00040
- Redden B, Maxted N, Furman B, Coyne C (2007) LENS Biodiversity. In: Yadav S S, McNeil D L, Stevenson P C (eds) *Lentil: an Ancient Crop for Modern Times*. Springer Heidelberg 11–22
- Risch S J, Andow D, Altieri M A (1983) Agroecosystem diversity and pest control: data, tentative conclusions and new directions. *Environ Entomol* 12:625–629. doi:10.1093/ee/12.3.625
- Rockström J, Steffen W, Noone K, Persson A, Chapin FS, Lambin E F, Lenton T M, Scheffer M, Folke C, Schellnhuber H J, Nykvist B, de Wit C A, Hughes T, van der Leeuw S, Rodhe H, Sörlin S, Snyder P K, Costanza R, Svedin U, Falkenmark M, Karlberg L, Corell R W, Fabry V J, Hansen J, Walker B, Liverman D, Richardson K, Crutzen P, Foley J A (2009) A safe operating space for humanity. *Nature* 461:472–475. doi:10.1038/nature11069
- Root R B (1973) Organization of plant-arthropod association in simple and diverse habitats: the fauna of collards (*Brassica oleracea*). *Ecol Monogr* 43:95–124. doi:10.2307/1942161
- Sandhu J S and Singh S (2007) History and Origin. In: Yadav S S, McNeil D L, Stevenson P C (eds) *Lentil: an Ancient Crop for Modern Times*. Springer Heidelberg 1–10

- Savage G P (1991) Lentils: a forgotten crop. *Outlook Agric* 109:112–20
- Saxena M C (2009) Plant morphology, anatomy and growth habit. In: Erskine W, Muehlbauer F J, Sarker A, Sharma B (eds) *The Lentil Botany, Production and Uses*. Cabi 34–46
- Schneider A, Huyghe C, Maleplate T, Labalette F, Peyronnet C, Carrouée B (2015) Rôle des légumineuses dans l’agriculture française. In : Schneider A, Huyghe C *Les légumineuses pour des systèmes agricoles et alimentaires durables*. Quae editions 11–77
- Sekhon H S, Singh G, Ram H (2007) Lentil-Based Cropping Systems. In: Yadav S S, McNeil D L, Stevenson P C (eds) *Lentil: an Ancient Crop for Modern Times*. Springer Heidelberg 107–126
- Seufert V, Ramankutty N, Foley J (2012) Comparing the yields of organic and conventional agriculture. *Nature* 485:229–233. doi:10.1038/nature11069
- Shah Z, Shah S H, Peoples M B, Herridge D F (1997) Surveys of N₂ fixation of lentil, mungbean, and blackgram in farmers’ fields in Northwest Frontier Province, Pakistan. *ICRISAT* 333:343
- Shearer G and Kohl D H (1986) N₂ fixation in field settings: estimates based on natural ¹⁵N abundance. *Australian J Plant Physiology* 699:756–13
- Shrestha R, Turner N C, Siddique K H M Turner D W (2006) Physiological and seed yield response to water deficits among lentil genotypes from diverse origins. *Australian Journal of Agricultural Research* 57:903–915.
- Shrestha R, Siddique K H M, Turner D W, Turner N C (2009) Breeding and Management to Minimize the Effects of Drought and Improve Water Use Efficiency. In: Erskine W, Muehlbauer F J, Sarker A, Sharma B (eds) *The Lentil Botany, Production and Uses*. Cabi 172–193
- Sidahmed M M, Jaber N S (2004) The design and testing of a cutter and feeder mechanism for the mechanical harvesting of lentils. *Bio Eng* 88:295–304. doi:10.1016/j.biosystemseng.2004.04.002
- Sinclair T R, De Wit C T (1975) Photosynthate and nitrogen requirements for seed production by various crops. *Sci* 565:567–189
- Singh T P (1977) Harvest index in lentil (*Lens culinaris* Medik.). *Euphytica* 26:833–839
- Slafer G A, Rawson H M (1994) Sensitivity of wheat phasic development to major environmental factors: A re-examination of some assumptions made by physiologists and modellers. *Aust J Plant Physiol* 21:393-426. doi: 10.1071/PP9940393
- Smil V (2002) Nitrogen and Food Production: Proteins for Human Diets. *J of the Hum Environ* 31:126–131. doi: 10.1579/0044-7447-31.2.126
- Solanki I S, Shyam S, Yadav S S, Bahl P N (2007) Varietal Adaptation, Participatory Breeding and Plant Type. In: Yadav S S, McNeil D L, Stevenson P C (eds) *Lentil: an Ancient Crop for Modern Times*. Springer Heidelberg 255–274
- Stefaniak T R, McPhee K E (2015) Lentil. In: De Ron Antonio M (ed) *Grain legumes*. Springer 111–140

- Subira J, Alvaroa F, Garcia del Moral L F, Royo C (2015) Breeding effects on the cultivar×environment interaction of durum wheat yield. *Europ J Agron* 68:78–88. doi :10.1016/j.eja.2015.04.009
- Szentesi A, Jermy T (1995) Predispersal seed predation in leguminous species: seed morphology and bruchid distribution. *Oikos* 73:23–32
- Tahvanainen JO, Root RB (1972) The influence of vegetational diversity on the population ecology of a specialized herbivore, *Phyllotreta cruciferae* (Coleoptera: Chrysomelidae). *Oecologia* 10:321–346. doi:10.1007/BF00345736
- Therond O, Duru M, Roger-Estrade J, Richard G (2017) A new analytical framework of farming system and agriculture model diversities 37:21. A review. *Agron Sustain Dev*. doi:s13593-017-0451-9
- Tosti G, Farneselli M, Benincasa P, Guiducci M (2016) Nitrogen fertilization strategies for organic wheat production: Crop yield and nitrate leaching. *Agron J* 108:770–781. doi:10.2134/agronj2015.0464
- Turay K K, McKenzie B A, Andrews M (1992) Effect of water stress and nitrogen on canopy development and radiation interception of lentil. *Proceedings Agronomy Society of New Zealand* 22:115–119
- Unkovich M, Herridge D, Peoples M, Cadisch G, Boddey R, Giller K, Alves B, Chalk P (2008) Measuring plant-associated nitrogen fixation in agricultural systems. Chapter 7: 15N–isotopic–general principles and 15N feeding. Australian Centre for International Agricultural Research (ACIAR) 125–131
- Vandermeer J H (1989) *The ecology of intercropping*. Cambridge, UK: Cambridge University Press
- Vermeulen S J, Campbell B M, Ingram J S I (2012) Climate Change and Food Systems. *Annu Rev Environ Resour* 37:195–222. doi:1543-5938/12/1121-0195\$20.00
- Viguié L, Bedoussac L, Journet E P, Justes E (2018a) Yield gap extended to marketable grain reveals the profitability of organic lentil-spring wheat intercrops. *Agron Sust Dev*, accepted
- Viguié L, Journet E P, Justes E, Bedoussac L (2018b) Species niche complementarity for nitrogen improves lentil-wheat intercrop grain production in organic farming. *Field Crops Res*, submitted
- Voisin A S, Guéguen J, Huyghe C, Jeuffroy MH, Magrini MB, Meynard JM, Pelzer E (2013) Legumes for feed, food, biomaterials and bioenergy in Europe: a review. *Agron Sust Dev* 34:361–380. doi:10.1007/s13593-013-0189-y
- Voisin A S and Gastal F (2015) Nutrition azotée et fonctionnement agrophysiologique spécifique des légumineuses. In : Schneider A, Huyghe C (Coord) *Les légumineuses pour des systèmes agricoles et alimentaires durables*. Quae editions 79–137
- Wang L, Gruber S, Claupein W (2012) Optimizing lentil-based mixed cropping with different companion crops and plant densities in terms of crop yield and weed control. *Org Agr* 2:79–87. doi:10.1007/s13165-012-0028-5
- Wang L, Gruber S, Claupein W (2013) Effect of sowing date and variety on yield and weed populations in a lentil–barley mixture. *J Agric Sci* 151:672–681. doi:10.1017/S0021859612000895

- Watson C A, Reckling M, Preissel S, Bachinger J, Bergkvist G, Kuhlman T, Lindström K, Nemecek T, Topp C F E, Vanhatalo A, Zander P, Murphy-Bokern D, Stoddard F L (2017) Grain legume production and use in European agricultural systems. *Adv in Agron* 144 :235–303. doi:10.1016/bs.agron.2017.03.003
- Whitehead S J, Summerfield R J, Muehlbauer F J, Coyne C J, Ellis R H, Wheeler T R (2000) Crop improvement and the accumulation and partitioning of biomass and nitrogen in lentil. *Crop Sci* 40:110–120. doi:10.2135/cropsci2000.401110x
- Willey R W, Osiru D (1972) Studies on mixtures of maize and beans (*Phaseolus vulgaris*) with particular reference to plant population. *J Agric Sci* 79:517–529
- Willey R W (1979) Intercropping—its importance and research needs. 1. Competition and yield advantages. *Field Crop Abstr* 32:1–10
- World Commission on Environment and Development (1987) *Our common future*, Brundtland report. Oxford University Press
- Yadav S S, McNeil D L, Andrews M, Chen C, Brand J, Singh G, Shivakumar B G, Gangaiah (2009) Soil Nutrient Management. In: Erskine W, Muehlbauer F J, Sarker A, Sharma B (eds) *The Lentil Botany, Production and Uses*. Cabi 194–212
- Yagmur M and Kaydan D (2006) Different intercrop arrangements with lentil and barley under dryland condition. *Pak J of Biol Sci* 10:1979–1922
- Yenish J P, Brand J, Pala M, Haddad A (2009) Weed Management In: Erskine W, Muehlbauer F J, Sarker A, Sharma B (eds) *The Lentil Botany, Production and Uses*. Cabi 326–342
- Yu Y, Stomph T J, Makowski D, Zhang L, Werf W (2016) A meta-analysis of relative crop yields in cereal/legume mixtures suggests options for management. *Field Crop Res* 269:279–198. doi:10.1016/j.fcr.2016.08.001
- Yus-Ramos R, Ventura D, Bensusan K, Coello-García P, György Z, Stojanova A (2014) Alien seed beetles (Coleoptera: Chrysomelidae: Bruchinae) in Europe. *Zootaxa* 3826:401–448. doi:10.11646/zootaxa.3826.3.1
- Zander P, Amjath-Babu T S, Preissel S, Reckling M, Bues A, Schläfke N, Kuhlman T, Bachinger J, Uthes S, Stoddard F, Murphy-Bokern D, Watson C (2016) Grain legume decline and potential recovery in European agriculture. *Agron Sustain Dev* 36–26. doi:10.1007/s13593-016-0365-y
- Zhu J, van der Werf W, Anten N P R, Vos J, Evers J B (2015) The contribution of phenotypic plasticity to complementary light capture in plant mixtures. *New Phyt* 207:1213–1222. doi:10.1111/nph.13416

List of figures

Fig. 1 Main models of agriculture (from 1 to 3b in blue) with farming systems identified based on their varying degrees of use of ecosystem services versus anthropogenic exogenous inputs (Y-axis) and connected to globalized food systems or local dynamics (Xaxis).	11
Fig. 2 (Left) sole cropped lentil and (middle) lentil-spring wheat intercrop at harvest, and (right) mechanical harvest of the intercrop.....	28
Fig. 3 Extension of the yield gap concept to the marketable yield.	30
Fig. 4 Yields and gross margins of lentil-spring wheat intercrops compared to those of sole cropped lentil.....	37
Fig. 5 Height, stem length and lodging of lentil intercropped with spring wheat compared to those of sole cropped lentil).	40
Fig. 6 Mean harvested yields of hand-harvested and mechanically harvested lentil in intercrop and sole crop, used to calculate mechanical harvest efficiency	42
Fig. 7 Yield gap analysis for lentil intercropped with spring wheat and sole cropped lentil... ..	44
Fig. 8 (a) Grain yield and (b) nitrogen accumulated in shoots of the total intercrop (IC) and sole crops (SC).....	56
Fig. 9 Partial land equivalent ratio of lentil in intercrops (LER_L) vs. partial land equivalent ratio of wheat in intercrops (LER_W)	58
Fig. 10 Grain yield vs. nitrogen accumulated in shoots for (a) wheat and (b) lentil in intercrops (IC) and sole crops (SC).	59

Fig. 11 For lentil, the intercrop (IC):sole crop (SC) ratio of the percentage of nitrogen (N) fixed from the air (%) vs. the IC:SC ratio of the amount of N fixed (kg ha ⁻¹)	60
Fig. 12 Grain protein concentrations of (a) wheat and (b) lentil in intercrops (IC) and sole crops (SC) as a function of grain yield	61
Fig. 13 Diagram explaining how species yield and yield components were measured or calculated for a) lentil and b) wheat.	75
Fig. 14 Lentil grain yield as a function of the wheat grain yield in intercrop and sole crops grain yield.	77
Fig. 15 Effect of the year on interspecific interaction indices of lentil and wheat for yield components in IC100%L:33%W	79
Fig. 16 Effect of wheat density on interspecific interaction indices of lentil and wheat for yield components.....	82
Fig. 17 Effect of lentil cultivar on interspecific interaction indices of lentil and wheat for yield components.....	84
Fig. 18 Effect of intercrops on weed abundance.....	100

List of tables

Table 1 Intercrop and sole crop treatments performed in the field in 2015 and 2016..... 51

Table 2 Effect of species densities and lentil cultivar on intercropped lentil and wheat grain yields averaged over both experimental years 57

Table 3 Field treatments performed in 2015 and 2016..... 72

Table 4 Correlation table for the lentil or wheat yield components and yield per m² according to the cropping system..... 80

Abstract

Author: Viguier Loïc

PHD directors: Laurent Bedoussac, Etienne-Pascal Journet and Eric Justes

Title: Analysis of the agronomic and economic performances of lentil-spring wheat intercrops in organic farming

Place and date of the defence: Toulouse, July 12th 2018

Lentil (*Lens culinaris* Med.) is an important component of the human diet in the world, but in the meantime, Europe produces only 26% of the lentils it consumes. This is partly due to strong agronomic weaknesses that reduce yield such as lodging, bruchid beetles and weeds, especially in organic farming. Intercropping, the simultaneous growing of two or more species in the same field is tested here as an option to reduce these drawbacks and develop organic lentil production.

The aims of this thesis were to (1) assess the potential of lentil-spring wheat intercrops to produce organic lentil, (2) understand the mechanisms that explain their performances, and (3) evaluate the profitability of such intercrops. A two-year field experiment was carried out in southwestern France in 2015 and 2016 under organic farming rules. Four lentil and two wheat cultivars were grown as sole crops and intercrops in multiple additive and substitutive designs.

Our results showed that the total intercrop attainable grain yield was higher than the mean of sole crops. Yet, lentil yield in intercrop was lower than in sole crop as the result of a strong competition for resources from wheat in early lentil growth stages reducing the number of branches per plant of lentil. This led to lower gross margins of intercrops.

However, lentil lodging was strongly reduced in intercrops thus its mechanical harvest efficiency increased. This led to similar mechanically harvested yields of lentil in intercrop and sole crop. Consequently, after mechanical harvest and grain cleaning, the marketable gross margin of intercrops was higher than that of sole crops.

Our results suggest that (1) intercrop had no effect on bruchids, (2) the most effective intercrop is when lentil is at sole crop density and wheat at 15-20%, (3) intercrop performance is due to complementary use of N pools through legume N₂ fixation and (4) the intensity of interspecific interactions depends on year, wheat density and genotypes.

Our work indicates that lentil-spring wheat intercrop can develop organic lentil production but a better understanding of Genotype x Environment x Cropping system interactions may be useful to design optimized managements.

Key words: Attainable Yield, Yield Gap, Actual Yield, Mechanical Harvest Efficiency, Lodging, Bruchid Beetle, Marketable Yield, Gross Margin, Resource Acquisition, Competitions, Nitrogen, Nitrogen Fixation, Grain Protein, Complementarity, Yield Components, Interspecific, Intraspecific

Résumé

Auteur : Viguier Loïc

Directeurs de thèse : Laurent Bedoussac, Etienne-Pascal Journet and Eric Justes

Titre : Analyse des performances agronomiques et économiques des associations de lentille et blé de printemps en agriculture biologique

Lieu et date de la défense : Toulouse, le 12 Juillet 2018

La lentille (*Lens culinaris* Med.) est une composante importante des régimes alimentaires de nombreuses populations à travers le monde mais sa consommation en Europe est relativement faible. L'Europe produit seulement 26% de sa consommation de lentille et ce déficit est en partie causé par d'importants verrous agronomiques comme la verse, les bruches et la compétition des adventices qui réduisent ses rendements, notamment en agriculture biologique. Les associations de cultures, définies comme la culture simultanée d'au moins deux espèces différentes sur une même surface pendant une durée significative, sont considérées comme une option pour lever ces verrous agronomiques et ainsi développer la production de lentille en agriculture biologique.

Les objectifs de cette thèse étaient de (1) évaluer le potentiel des associations de lentille et de blé de printemps pour produire de la lentille en conditions d'agriculture biologique et (2) comprendre les principaux mécanismes sous-jacents à la performance des associations. Des essais agronomiques ont été mis en place en 2015 et 2016 en conditions d'agriculture biologique. Quatre variétés de lentille et de blé de printemps ont été conduites en culture pures et en plusieurs associations de type substitutif et additif.

Nos résultats montrent que le rendement moyen des associations avant récolte mécanique était plus élevé que le rendement moyen des cultures pures. Néanmoins, le rendement de lentille en association était inférieur à celui de la lentille en culture pure en raison d'une compétition forte et précoce du blé pour les ressources qui a causé la diminution nombre de ramifications par plante de la lentille. Le prix de la lentille étant environ quatre fois plus élevé que celui du blé, la marge brute des associations avant récolte était inférieure à celle de la lentille en culture pure.

Cependant, la verse de la lentille a été fortement réduite en association, entraînant une augmentation de l'efficacité de sa récolte mécanique. En conséquence les rendements de lentille issus de la récolte mécanique se sont avérés similaires en association et en culture pure. Enfin, après tri et nettoyage des graines, la marge brute des associations sur le rendement commercialisable était supérieure à celle des cultures pures.

Nos résultats montrent que (1) les associations n'ont pas eu d'effet sur le taux de bruchage des lentilles, (2) l'association la plus performante est constituée de lentille à densité équivalente à la culture pure dans laquelle on ajoute 15-20% de blé, (3) la performance des associations est due à une utilisation complémentaire de l'azote rendue possible par la fixation symbiotique de l'azote par la lentille et (4) l'intensité des compétitions entre espèces dépendent de l'année, de la densité de blé et des génotypes.

En conclusion, nos travaux indiquent que les associations de lentille et de blé de printemps peuvent permettre de développer la production de lentille en agriculture biologique mais qu'une meilleure compréhension des interactions de type génotype x environnement x conduite pourrait permettre de mettre au point des couverts encore plus performants.

Mot clés : Rendement Atteignable, Ecart de Rendement, Rendement sur Pied, Efficacité de Récolte Mécanique, Verse, Bruche, Rendement Commercialisable, Marge Brute, Acquisition de Ressources, Compétition, Azote, Fixation Azote, Protéine, Complémentarité, Composantes du Rendement, Interspécifique, Intraspécifique