YIELD AND NUTRITIVE VALUE OF ALFALFA (*Medicago sativa* L.) IN AN OLIVE (*Olea europaea* L.) ALLEY-CROPPING PRACTICE

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Introduction

Among silvoarable systems, alley-cropping is a practice where annual or perennial crops are intercropped between wide rows of trees. In Italy, the most cultivated fruit tree is olive (*Olea europaea* L.), in fact its cultivated area represents 60% of orchards, with 1.1 millions of hectares. Eichhorn *et al.* (2006) identified an area of 20.000 ha in central Italy (Umbria and Lazio Region) of silvoarable olive grove practice.
In Tuscany traditionally, the soil of olive grove is covered by a low-productive natural pasture, usually not grazed, until the early-summer period when surface tillage is performed in order to decrease water competition between herbaceous plants and trees. Intercropping tree crops with legumes, and in particular perennials legume species, is a way to improve sustainability of Mediterranean silvoarable olive grove: increasing the Nitrogen (N) content in the soil by the N fixation (Anglade *et al.*, 2015), reducing the use of inorganic N and enhancing soil cover protection by plant (Vallebona *et al.*, 2016).
The most important legume crop in the Mediterranean basin is alfalfa (*Medicago sativa* L.), a perennial forage species able to produce a plentiful biomass even under rainfed conditions. There is an interest in alfalfa cultivation in alley-cropping systems as hay crop but also for grazing use. Facilitation and limitation in an agroforestry system mainly depend on functional tree group (Rivest *et al.*, 2013), soil fertility level (Moreno 2008) and climate condition (Moreno *et al.*, 2007). Tree and herbaceous layer interact for resources as light, water and soil nutrient.
A field experiment was carried out in order to investigate the suitability of alfalfa meadow in an alley-cropping silvoarable system under Mediterranean conditions for two consecutive years in a rainfed mature olive grove.

Material and methods

In the 2014 and 2015 a field experiment was conducted in Manciano (42° 32.512', 11° 26.684') in south Tuscany. The field trial was established in a 70 year-old rainfed olive grove and control plots were carried out in the bordering field. The display layout of the olive grove is 5 x 10 m and it is located on a uniform terrain, with a slope of 9% and South-East orientation. Tree rows are planted according to the terrain aspect (NW-SE). The central portion of two side by side alleys (about 7 m large) and the bordering control field were tilled (chisel ploughing, disk harrowing twice) at the end of winter season 2014. Then, alfalfa (Cultivar Messe) was sown (seed dose 40 kg ha^{-1}) inside the olive grove and outside. The experimental design was a randomize block with 3 replicates, with plot sized 2.4 x 10 m. Analysed treatments were: Open field (OF), Central part (CA), North-facing side (NA) and South-facing side (SA) of the alleys. In 2014 and 2015 alfalfa was harvested when the 10% of the open-grown crop was at flowering stage.
During the establishment year, the management of the alfalfa meadow was conducted under a four-harvest cycle. Harvesting dates were 23 June (H1), 30 July (H2), 5 September (H3) and 20 October (H4). In 2015 the crop was harvested six times: 30 April (H5), 28 May (H6), 23 June (H7), 1 August (H8), 16 September (H9) and 22 October (H10).
At each harvest time, an area of $1m^2$ per plot was sampled, cutting the herbage at 3 cm fixed height from the ground. Alfalfa biomass subsamples were placed in a forced-draft oven at a temperature of 60 °C in order to determine the dry matter (DM) content and the Above-Ground Biomass Yield (AGBY). Chemical analyses for valuing forage quality were conducted on milled samples (1mm), determining Crude Protein (CP), Neutral Detergent Fiber (NDF), Acid Detergent Fiber (ADF) and Acid Detergent Lignin (ADL) content. A split-plot ANOVA was used to investigate differences between alfalfa DM production among treatments (T) and harvest time (H) considered as subplot factor.
The under-canopy available total light transmittance was estimated from hemispherical photos, taken in all the alley-cropping system plots, according to a regular grid of 144 points at increasing distance from the tree rows (0, 1, 3, 5 m).

The images were processed by Gap Light Analyser 2.0 free software. Spatialization of point data was performed by Inverse Distance Weighting (IDW).

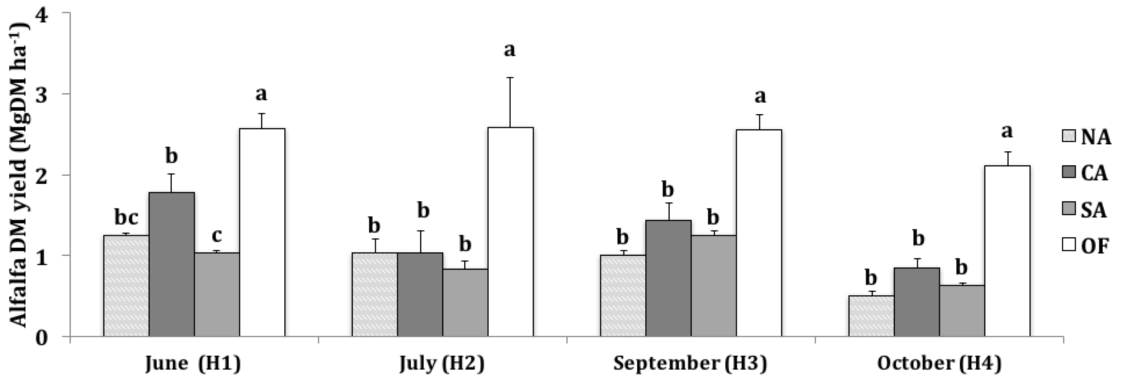
Results

During 2014, cumulative rainfall from seeding (March 20th) to the end of the harvest period (October 20th) was about 580 mm with rainfall over the monthly climatological mean in June, July and August. In 2015, for the same period, cumulative rainfall was 392 mm, in line with the long-term average. Average mean temperature of March-October period was about 19.2 and 19.9 °C in 2014 and 2015, respectively. In 2014 no drought period was observed, while in 2015 May, July and September can be considered dry (Bagnouls and Gaussen, 1957).
The annual alfalfa yield was lower in 2014 (establishment year) in all treatments than in 2015 (**Table 1**). AGBY was significantly higher in OF, 9.81 and 12.14 Mg DM ha^{-1} yr^{-1} in 2014 and 2015, respectively. During 2014, no significant differences were observed among the plots included in the olive grove. On average, from the first to the second year, AGBY in NA, CA and SA increased by 18, 19 and 27%, respectively. In both years, alfalfa yield was affected by treatment and harvest time while interaction between factors was not significant.

Table 1: Mean total annual dry matter yield of alfalfa grown in open field (OF) and inside an olive grove (North (NA), Central (CA) and South (SA) of alley) in Manciano, Italy.

	Year 2014			***Year 2015***		
Treatment	**Annual yield (Mg DM ha^{-1})**	**SE§**		**Annual yield (Mg DM ha^{-1})**	**SE**	
OF	9.81	± 0.64	**a**	12.14	± 0.34	**a**
NA	3.79	± 0.28	**b**	4.48	± 0.40	**c**
CA	5.12	± 0.80	**b**	6.51	± 1.20	**b**
SA	3.75	± 0.20	**b**	4.49	± 0.54	**c**

Same letters indicate that treatments are not different at the 0.05 probability level according to Tukey's HSD test
§ Standard Error of the Mean



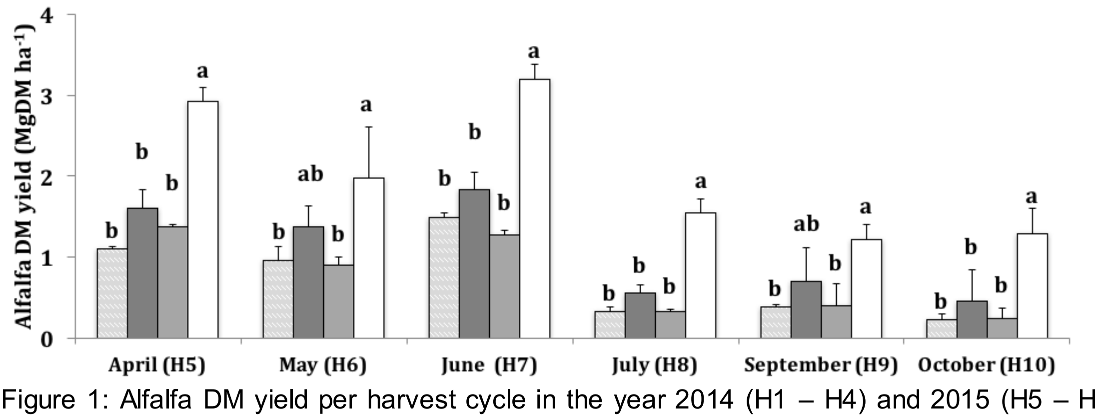


Figure 1: Alfalfa DM yield per harvest cycle in the year 2014 (H1 – H4) and 2015 (H5 – H10) among treatments: open field (OF), north-facing side of the alleys (NA), central part of the alleys (CA) and south-facing side of the alleys (SA) in Manciano, Italy. Same letters indicate that treatments are not different at the 0.05 probability level according to Tukey's HSD Test. Vertical bars represent standard errors.

In the first year, at each harvest time AGBY was higher in OF than NA, CA and SA (**Figure 1**). The yield reduction in CA was about 31, 60, 40 and 60% compared to OF in H1, H2, H3 and H4, respectively. The highest reduction levels were recorded for NA and SA in H4, about 75 and 70%, respectively compared to OF.
In 2015, AGBY in OF was significantly higher than in alley treatment at each harvest time with exception of H6 and H9 (**Figure 1**). From OF to CA, we observed a lower AGBY reduction than from OF to NA and SA at each harvest time. ABGY in CA was about 45, 31, 42, 65, 42, 65% lower than OF, in H5, H6, H7, H8, H9 and H10, respectively. The lower AGBY observed in the alley treatments was determined by the lower available light transmittance (ALT) measured in the olive grove. **Figure 2** shows the direct linear correlation between annual ABGY and ALT in both years, with *r*-Pearson value of about 0.92.
Regarding forage quality, in the first year, the CP content of alfalfa collected in OF was significantly lower than NA and SA in the last two harvest time H3 and H4, and in the latter it was also significantly lower than CA. While, NDF and ADF contents in OF were higher than NA, CA and SA in H3 and H4. ADL content was higher in OF than NA, CA and SA only in H3.
In 2015, no significant differences were observed for CP and ADL among treatments and harvest times. The lowest NDF content was observed in NA at the first harvest time of 2015. In the same year ADF content in OF significantly increased compared to the alley treatments in the summer period (H7 and H8).

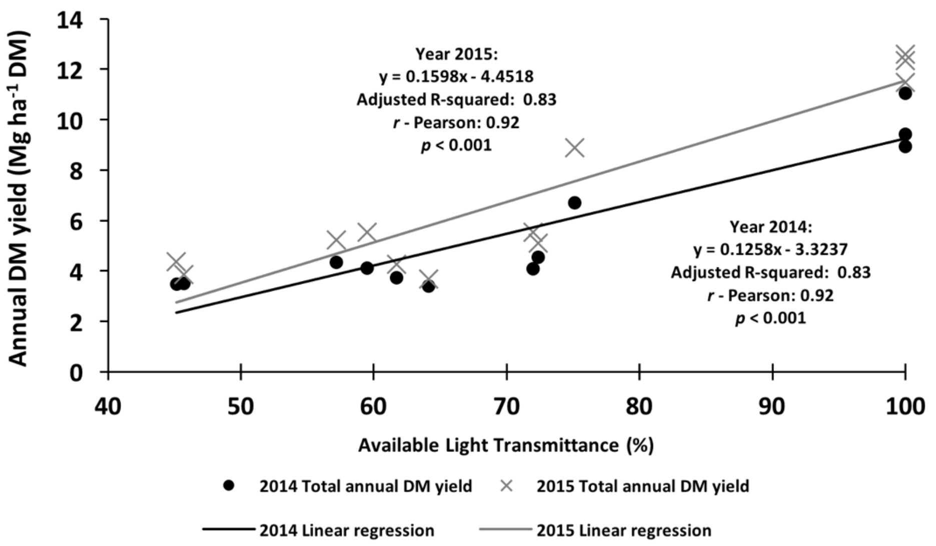


Figure 2: Relationship between available light transmittance and alfalfa yield of each plot in the growing season 2014 and 2015

Discussion

This work investigated the forage biomass production in terms of yield and quality of alfalfa cultivated in a typical olive grove of south Tuscany. The grass-tree interaction was investigated in order to evaluate the suitability of alfalfa in a silvoarable practice. Our results provide evidence of a lower alfalfa productivity in an olive alley-cropping system. The negative effect of alfalfa-tree interaction was already reported in a 20 years-old black walnut (*Juglans nigra* L.) alley-cropping plantation situated in North America (McGraw *et al*., 2008), and also in a 7x7 m layout radiata pines (*Pinus radiata* D. Don) agroforestry trial in New Zealand (Varella *et al*., 2011). The alfalfa yields of border alley sides (NA and SA) were lower than the central part of the alley (CA) showing a higher competition with trees for resources. The highest reduction was recorded in both years in summer period harvests (H2 and H7) and in the last harvests before overwintering (H4 and H10). This probably was caused by the competition for water in summer period when precipitations were not abundant. Nevertheless, the low light availability in the olive groves is likely to be the main limiting factor for biomass accumulation in alfalfa meadow. Several authors reported a negative effect of reduced light availability on legumes growth in agroforestry systems (Moreno *et al*. 2007; Gea-Izquierdo *et al*., 2009; Lopez-Carrasco *et al*., 2015). Moreover, McGraw *et al*. (2008) observed a biomass reduction of alfalfa according to distance from the trees in an alley-cropping practice.

Nutritive value of alfalfa was not negatively affected by tree competition as no reduction of crude protein content was observed in the alley-cropping treatments. As previously reported (McGraw *et al*., 2008), low variability was found in CP, NDF and ADF concentrations between the alley-cropped and open-grown alfalfa. In our study, also ADL showed few difference among treatment, regardless the harvest time.
Concluding, alfalfa biomass reduction in the olive grove mainly depended on light availability. Anyway, further studies are needed in order to investigate the complex of interactions between tree and herbaceous crop in agroforestry system, addressing the possible competition for water and nutrients. Tree management, as pruning, can enhance the meadow productivity and tree study approach is necessary to investigate the effect of alfalfa presence in wide olive tree plantation productivity. Moreover, further investigation should also address the presence of anti-nutritional factors (i.e. saponins and tannins) in alfalfa grown under agroforestry systems in order to evaluate the suitability of alfalfa to different utilizations.

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