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Nutritional composition of spineless safflower (*Carthamus tinctorius* L. var. *inermis* Schweinf.) grown at different levels of nitrogen fertilization.

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ABSTRACT

Safflower is traditionally grown all over the world for its seeds and the dyeing properties of the flowers, so it is commonly used in food industry for coloring and flavoring foods, in pigment industry and in the oil production industry. Some studies have shown that safflower could be grown as a winter crop in areas with mild temperatures or as spring crop in cooler areas. To gain insight about the potential of safflower as alternative forage crop within crop rotation plans of agro-zootechnical farms, our study was aimed to assess how N-fertilization (0, 35, 70 and 105 kg ha^{-1}), affected yields and nutritional value of spineless safflower grown under Central-Italy conditions, typical of the Tyrrhenian costal area. In terms of dry matter yield, the biomass production showed a nitrogen-dependence passing from 1.80 tons per hectare, as a mean without nitrogen supplementation, to 2.71 tons per hectare fertilizing at 105 kgN ha^{-1}. As far as the crude protein content is concerned, nitrogen fertilization has had a significant effect on nitrogen accumulation of safflower passing from about 11% at lower level up to more than 17% for the highest level tested. In general, as far as fat, fibers, ashes and no cell wall CHO, there were not significant differences among treatments apart for an unexpected high value for lignin and ADF corresponding at the medium level of nitrogen fertilization tested. These findings showed that N-fertilization played an important role in the productivity of safflower and, particularly, on CP content of forage. Spineless safflower showed an interesting potential to be grown for ruminants feeding purposes in Mediterranean EU countries, but further trials are needed to fully evaluate safflower performances either as hay or as silage and to find the best conditions (time of sowing, seed rate, varieties implied) giving higher forage yields.

INTRODUCTION

Safflower (*Carthamus tinctorius* L.) is a plant belonging to the Asteraceae family. It grows wild in the Middle East and also along the Mediterranean French coast. Because of its tolerance to droughts, it is cultivated in many hot regions as an oilseed, birdseed or for its flowers which are used as dye sources and for medicinal purposes (Koutroubasa and Papakosta, 2010). It is a minor crop today, with about 600,000 tons being commercially produced in more than sixty countries worldwide. India, United States, and Mexico are the main producers, followed by Ethiopia, Kazakhstan, China, Argentina and Australia. The importance of safflower as oilseed crop has increased in recent years, especially with the increased interest in the production of biofuels (Dordas and Sioulas, 2008). Interesting is also the potential use in medicine for treatment of hyperlipemia, coronary heart disease (CHD) and for the capacity to enhance microcirculation because of the high content of linoleic acid (70–87%), tocopherols, phytosterols and phenols compounds of the seeds (Nogala-Kalucka *et al*., 2010). From an agronomical point of view, safflower shows a good drought, salt and heat tolerance (Zaman and Maiti, 1990; Welti *et al*., 1998), and its deep tap root enables it to use nutrients below the root zone of cereal crops, potentially reducing the rate of nitrogen (N) application. As a matter of fact, cropping safflower in rotation with other species may have environmental benefits as well as a positive role in saving the costs of nitrogen-fertilization (Yau and Ryan, 2010). Growing safflower after a fertilized crop may remove N from the lower part of the soil profile reducing the elution risk of the nutrient from the soil to the ground waters. Recently, spineless cultivars of safflower have been evaluated for forage production (Leshem *et al*., 2001; Landau *et al*., 2004; 2005). Moreover, safflower seed or decorticated meal (about 40% protein) may be of interest for protein supplementation in beef cattle (Bottger *et al*., 2002; Scholljegerdes *et al*., 2004; Bolte *et al*., 2002) and sheep (Kott *et al*., 2003) diets. Pre-partum supplementation with safflower seeds high in either linoleate or oleate has increased the subsequent conception rates in primiparous beef cows (Lammoglia *et al*., 1997). Safflower as forage has also been reported to improve fertility in Canadian ewes, in comparison to alfalfa-grass hay (Stanford *et al*., 2001). Safflower is a very adaptable crop that can be sown in winter or spring in both in dry and irrigated cropping systems and has the potential of lengthening the duration of lush green pastures under arid conditions. Safflower based pastures appear adequate for growing ruminants having moderate nutritional requirements (Landau *et al*., 2005). Since safflower is an under-studied crop, the relationship between safflower and N is poorly understood, with even contrasting reports on the optimal rate of N fertilization (Knowles and Miller, 1960; Cazzato *et al*., 1997). The high yields which can be obtained growing safflower as a fodder crop (Leshem *et al*., 2001) and the great adaptability of this plant to arid and semi-arid environments, may provide many growers with an alternative crop option in cropping systems of the Mediterranean areas (Weinberg *et al*., 2002; Bar-Tal *et al*., 2008; Ghamarnia and Sepehri, 2010). The purpose of the present study was to assess the effect of nitrogen fertilization on yield and nutritional characteristic of harmless safflower intended for ruminant feeding.

MATERIAL AND METHODS

Experimental design

The study was carried out in Viterbo, Central Italy (42° 26’ N, 12° 04’ E, altitude 310 m a.s.l), from 2008 to 2009 on a clayey soil (45.20% clay, 24.50% silt, 30.08 sand, pH 7.33, 0-30 cm depth). Spineless safflower was sown in November 2008 and was grown under rainfed conditions. Total precipitation within the growing season (November 2008 – May 2009) was 657.6 mm (data obtained by the weather station of the Department of Crop Production). The experimental design was a randomized block with three replicates. The plots’ size (18 m x 3 m) was 54 m^2 and plant density was adjusted to 45 m^{-2}. Four nitrogen fertilization levels were applied at the beginning of the stem elongation stage (March 2009): N_0 = 0 kg ha^{-1} (residual soil fertility, N tot. 0.16%, O.M. 2.13%), N_1 = 35 kg N ha^{-1}, N_2 = 70 kg N ha^{-1}, N_3 = 105 kg N ha^{-1}.

Sample pre-treatment

Plants were mown at the end of the branching stage, when the young buds appeared, and were left in the field for a 2-days wilting treatment in view of a possible use for ensiling trials. On each plot, a representative sample (approx. 6 kg) of fresh and wilted plant material was collected for the determination of the yields and chemical analysis. On wilted plant material (chopped at 2-3 cm length) pH was measured by means of a Crison micropH 2002 pH-meter (Crison, Esp). All dry plant samples (at 65°C for 48 h in a forced air oven) were ground (Retsch Müller, Germany) to pass a 1 mm screen and stored in hermetic jars for the following analyses.

Chemical analysis

Dried samples were analyzed for crude protein (CP) by distillation Kjeldal method, lipids (EE) by ether extraction, Crude Fiber (CFom) and ashes (ASH) according respectively to AOAC Official Methods 984.13 (A-D), 920.39, 978.10 and 942.05 (AOAC, 2006). Also, Neutral Detergent Fiber (aNDFom) was assayed according to the method of Mertens (2002), Acid Detergent Fiber (ADF) was determined according to AOAC Official Method 973.18 (A-D) (AOAC, 2006) and lignin (ADL) was determined by sulphuric acid method (Robertson and Van Soest, 1981; AOAC Official Method 973.18, A-D, 2006). Non-fiber carbohydrates (NFC) were calculated as [100 − (NDF + CP + EE + ASH)].

Statistical analysis

All data were subjected to the analysis of variance using STATISTICA 7.0 (StatSoft, Inc, USA). Differences were compared by the Fischer LSD post-hoc test and declared significant at $p<0.05$.

RESULTS

At mowing, the biomass production (wet weight) was significantly affected ($p<0.01$) by nitrogen application passing from about 10 tons per hectare to more than 18 tons per hectare, even if no statistical differences ($p>0.05$) were observed fertilizing either with 35 or 70 kg of nitrogen per ha. Also, a direct relationship was observed between N-fertilization and DM yield (from N_0 = 1.80 ± 0.30 t ha^{-1} to N_3 = 2,71 ± 0.20 t ha^{-1}; $p<0.01$) (Tab. 1). Plants showed a mean DM content of 16.5% just after mowing even though the N_2 and N_3 fertilized ones had a higher ($P<0.05$) moisture content (85±4% and 85±3%) compared to the control (81±2%). On wilted plant material, all N-treated samples showed a significant ($p<0.01$) lower DM value than the control (Tab. 1).

Table 1. Yields, moisture content at mowing and DM content of wilted safflower grown under different level of N-fertilization. ^{a,b,c} $p<0.05$, ^{A,B} $p<0.01$.

N-level	Yield (t ha^{-1})*	Yield (t ha^{-1})**	Moisture at mowing**	DM after wilting
N_0	$9.8±2.0^{Bc}$	$1.8±0.3^{Bc}$	$0.81±0.02^{b}$	$0.36±0.01^{A}$
N_1	$13.9±3.3^{b}$	$2.3±0.5^{ab}$	$0.83±0.02^{ab}$	$0.31±0.01^{B}$
N_2	$14.6±3.2^{b}$	$2.1±0.5^{bc}$	$0.85±0.04^{a}$	$0.30±0.02^{B}$
N_3	$18.1±2.4^{Aa}$	$2.7±0.2^{Aa}$	$0.85±0.03^{a}$	$0.32±0.01^{B}$

*Data (mean±s.d.) are expressed on wet weigh (WW) basis;
**Data (mean±s.d.) are expressed on dry matter (DM) basis;

In view of ensiling trials, pH was determined on wilted plant samples. Overall, the highest N-treated plants (N_2 and N_3) showed a significant ($p<0.01$) higher pH values than the N_1 and N_0 treated ones (Fig. 1). A slight significant difference ($p<0.05$), was also observed between treatments N_2 and N_3 (Fig. 1). Looking at safflower as forage intended for ruminant feeding, no differences were observed among treatments as for the nutritional parameters EE, CF_{om}, $aNDF_{om}$, NFC and ASH and only for the N_2 treatment were recorded a higher content ($p<0.05$) of ADF and ADL (Tab. 2). Also, the N_3 treated plants showed a lower, though not significant ($p>0.05$), level of $aNDF_{om}$ (42.9 ± 1.9%) compared to other treatments and control. At the same manner, a moderate decrease in the NFC content was recorded for the N_3 samples (Tab. 2).

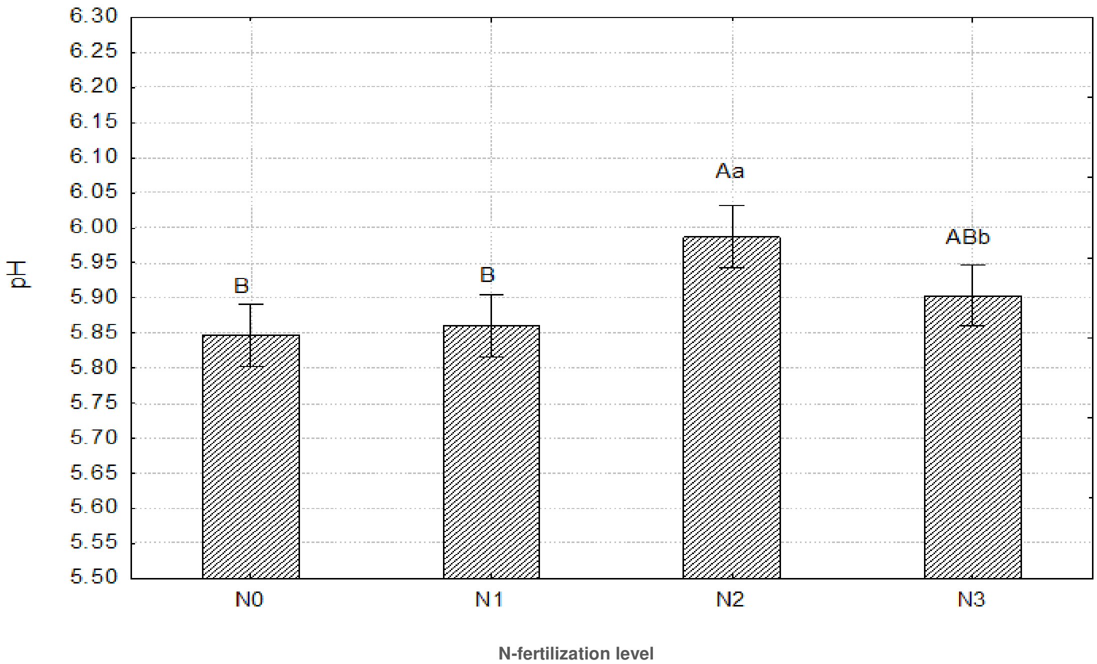

Figure 1. pH of 2-days wilted safflower plants as affected by the nitrogen fertilization. Data are expressed as Lsmean±SEM. ^{A,B} p<0.01; ^{a,b} p<0.05.

Table 2. Proximate chemical composition of spineless safflower grown under different level of N-fertilization. ^{a,b} p<0.05; ^{,A,B,C} p<0.01.

N-level	EE	CP	CF_{om}	$aNDF_{om}$	ADF	ADL	ASH	NFC
N_0	1.3±0.2	11.27±0.54^C	43.8±4.7	45.9±1.7	34.4±1.4^b	9.4±1.6^b	13.2±0.6	28.3 ± 1.5
N_1	1.4±0.3	15.31±0.49^B	42.1±3.5	44.7±3.9	33.7±1.8^b	7.4±1.9^b	12.7±0.4	25.8 ± 4.6
N_2	1.3±0.2	16.02±1.64^{ABb}	39.8±3.0	45.1±2.5	35.4±2.3^a	11.7±1.8^a	12.7±1.1	24.8 ± 3.3
N_3	1.4±0.3	17.50±2.27^{Aa}	41.2 ±2.7	42.9±1.9	33.1±2.7^b	9.6± 2.4^b	12.4±0.3	25.8 ± 0.6

*Data (mean±SD) are expressed as % on dry matter (DM) basis.

As far as the crude protein content of spineless safflower is concerned, the N-treated plants showed a higher content compared to the control passing from about 11% DM to more than 17% (Tab. 2). At the low (N_1) and medium (N_2) level of N-fertilization were not observed significant difference (p>0.05) in the CP content.

DISCUSSION

As expected, the fresh biomass production of spineless safflower was affected by the nitrogen fertilization due to a combined effect of DM and water retention increase. In terms of DM yield, the highest value obtained (2.7±0.2 t ha^{-1}) fertilizing at 105 kgN ha^{-1} was lower than the yields reported by others (Smith, 1996; Leshem *et al.*, 2000; Landau *et al.*, 2005, Weimberg *et al.*, 2007). In most cases, however, the trials were designed specifically for forage production with a seed rate higher the one we tested (about 20 kg ha^{-1}) which was originally adopted for seed and dye production and only secondarily for biomass production. Most probably, the low plant density used in our study was the main factor effecting the biomass production. As suggested by results found in literature (Weinberg *et al.*, 2007), an increased plant density (up to a seed rate of 45 kg ha^{1-}) combined with a higher nitrogen fertilization, could further improve the biomass production reaching DM yields comparable with those of many winter cereals (Smith, 1996). The water content of fresh safflower mowed at the budding stage, was affected by N-fertilization and in general the moisture data were found to be too high to guarantee correct ensiling. Wilting safflower forage for one or more days is very effective at the early stage of harvesting, as this practice greatly increases DM, and, at the same time, reduces the intensive fermentation and protmeaneolysis processes of the silage. The 2-days wilting treatment performed in our study, gave satisfactory results in terms of DM content, especially for plants grown in N-fertilized plots having a mean DM content ranging from 30% to 32%. Comparing safflower ensiled either at 41% DM or 29% DM, Weinberg *et al.* (2002) observed a faster pH decrease for 29% DM than the for driest forage regardless the inoculation of *Lactobacillus plantarum* (3.3 x 105 UFC g^{-1}). These facts suggest that the 2-days wilting treatment of safflower up to about 30% DM, may be considered satisfactory to give good ensiling kinetics. If harvesting is performed at the flowering stage, wilting might be unnecessary (Corleto *et al*., 2008). On the other hand, a recent research on the *in vitro* organic matter digestibility (IVOMD) of safflower (Peiretti, 2009) showed a linear reduction in gas production with advancing of stage maturity probably reflecting a decrease in nutritive value (from 805 to 588 g/kg OM respectively for late vegetative and early flowering stages, with a mean decrease of 3.3 g kg^{-1} OM day^{-1}). Overall, the nutritional characteristics were in line with data reported in literature for safflower (Landau *et al*., 2004; Weinberg *et al*., 2007) and are substantially comparable as for CP, lipids, NDF and ADF, with data found in literature (Martillotti *et al*., 1996) for many common forage cereals (bromegrass resque, italian ryegrass, tall fesque, orchardgrass) and some forage legumes (alfalfa, sainfoin, squarroso clover and vetch). In our study, the nitrogen fertilization had a slight effect on nutritional characteristics, apart the CP content that was strongly nitrogen-dependent. In similar circumstances, Weinberg *et al.* (2007) did not find any statistically significant effect on NDF content of safflower grown under different N-fertilization treatments (0 *vs*. 300 kg N ha^{-1}) while a significant effect was found on the ADF content. Safflower is also known to contain a moderate amount of polyphenolic compounds, including tannins, which may improve the nutritional value of this species, especially as far as the protein metabolization in ruminants (Grabber and Coblentz, 2009). As a matter of fact, moderate amounts of tannins in forages are able to partially protect dietary protein from microbial degradation in the rumen, increasing the absorption of amino acids from the small intestine (Waghorn, 1996) and improving animal performances such as milk production, wool growth and ovulation rate (Min and Hart, 2002). Tannins together with a good content of non-fibrous carbohydrates, may explain the good palatability shown by safflower (Dajue and Mündel, 1996) as well as by some legume forages (Jones *et al*., 1976). Further studies are needed to evaluate this particular aspect having also environmental implication as far as the reduction of green house gases emission by livestock.

CONCLUSION

This preliminary results obtained indicate that the selection of spineless safflower used has a good nutritional value which may be partially improved by the nitrogen fertilization. Moreover, wilted safflower characteristics allow satisfactory preservation by ensiling. Further studies are in progress to fully evaluate, both from agronomical and nutritional points of view, the potential of this species as forage crop for integrated crop-livestock production systems in Central Italy.

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