Buffalo's Grazing of Native and Improved Sward Pasturelands in Protected Environments of Mediterranean European Union

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Abstract: Herbage of Mediterranean pastures in the last decade weakened the production with a consequence on livestock and on environmental landscape. Fertilizer application was used to improve soil fertility located in two pasturelands [Environment I (Gioi) and Environment II (Monterotondo)] of Mediterranean environment belonging to Campania and Latium districts, respectively. The effect of fertilizer applications on herbage was evaluated by grazing with buffalo heifers and tested with a similar folk bred in cattleshed. The effect of pastures improvement was assessed on undisturbed areas of sward herbage growth in protected cages. No variation in biochemical characteristics of topsoil was made by fertilizer application. The grass of sward was browsed by a group of 18 heifers during the period September 2005 to November 2006. The effect of treatment increased, in the period of pasture utilization March-June, the content of native pasture parameter: dry matter (53% at Environment I and 27% at Environment II), nutritive value of herbage (0.016 Milk Forage Unit at both environments), flora composition (mean over locations, Graminaceae, was higher 22.4 and Leguminosae and miscellaneous group both lower 15 point of percentage than the content of the flora of the native pasture) and weight body gain of buffalo heifers which browsed sward the improved pasture was higher 27.6% in Environment I and 26.8% Environment II than in native. The weight body gain of animal at grazing in comparison to those bred in cattleshed was lower 40% and 19.4% in native and 25.7% and 1.4% in improved of E I and E II pastures, respectively.

Key words: Buffalo heifers, Mediterranean pastures improvement, nutritive values of herbage, sward phytocoenoses composition.

1. Introduction

Herbage production of Mediterranean pasture depends on soil fertility and meteorological events occurred during development of the herbage in the sward. The intensification use of the Mediterranean pastures promoted high stocking rates with consequent negative effect on sward, flora composition, nutritive quality of the herbage and environmental landscape [1, 2].

The herbage production and the ecological function of the pastures are linked to development of agro-pastoral techniques of management able to recover the native biodiversity of pasturelands [3, 4]. In the last decade, intensive grazing activity stressed the sward causing overgrazing with a consequent reduction of biodiversity and herbage production [5, 6]. In order to restore the native biodiversity in pasturelands, various approaches have been proposed (fertilization, irrigation and reseeding) [7, 8]. Among them, fertilizer applications, for a short period was the practices mostly used for recovering biodiversity, herbage productivity and nutritive characteristics of native pastures in many Country of the world [9-14]. The effect of soil enrichment by chemical fertilizer increased soil fertility favouring germination of the

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indigenous seed-bank present in the sward and development of native characteristics of herbage pasturelands [15]. Furthermore, because the biodiversity existing in protected pasturelands of Mediterranean European Union territories (National Park, Sites with Community Importance and Zones with Special Protection) are protected by European law, the increase of soil fertility in topsoil represent useful agronomic practice available for recovering biodiversity without allows genetic contamination [3, 5, 6]. The other agronomic practices, like reseeding with exogenous ecotypes, because the application introduce foreign genotypes which may promotes genetic contamination of flora in the sward of UE protected rangelands, they are forbidden by UE law [16].

The experiments aimed to assess information on the effect of chemical application in two pastures of Mediterranean environments on herbage of sward (dynamics of biomass growing, phytocoenoses composition, dry matter yield, milk feeding value of herbage), on weight body gain of grazing young buffalo heifers and on biochemical properties of soil.

2. Materials and Methods

The experiments were established in two pasturelands located in southern Italy though the year 2006 and 2007. The native pasture referred as Environment I [Gioi, longitude 15°13' E; latitude 40° 17' N and 684 m above sea level, (msl)] and Environment II (Monterotondo, longitude 12°57' E, latitude 42°03' N and 165 msl) belong to Zones with Special Protection of the UE and are located in Campania and Latium regions of southern Italy, respectively [17]. The environment I and environment II, further in the text, tables and figures will refer as E I and E II, respectively. The meteorological weather characteristics of two years of rainfall, mean temperature, evapotranspiration (Eto) determined by Class A water pan and solar radiation by radiometer, occurred in the areas of E I and in E II across the years of experiment, were reported in Fig. 1.

The native and improved sward pasturelands were grazed by 18 female buffalo heifers of about 6-8 months old and removed from browsed areas when the animals were near the age of puberty (about 24 months old) while another group of 18 heifers for the same period was intensively bred in cattleshad and fed with unifeed.

The environments of pasturelands showed yearly mean of rainfall (Fig. 1a), mean temperature (Fig. 1b), Eto (Fig. 1c) and radiation (Fig. 1d) 133 mm of rain lower and 7 °C, 653 mm of water and 251 MJ m² day higher in E I than E II, respectively. The amount of rainfall felt during the phonological stage of flora development in the months March to May was 138 mm in E I and 209 mm in E II and the water lost by Eto was 452 and 305 mm in E I and E II, respectively. In the following months, from June to September, the values of weather parameters temperature, Eto and solar radiation were increased and rainfall was reduced in E I than E II (37.8%, 37.5%, 5.2% and 35.7%) (Fig. 1). The solar radiation value (mean over environments) in the E I and E II decreased from August to December with similar trend passing from 706 to 165 MJ m² day while the mean temperature and ETo of E I in August to December were about 9 °C and 125 mm of evaporated water higher than E II (Figs. 1b-1d).

The sward of pastures, in middle October of the year 2005 and 2006 were fertilised with nitrogen and phosphorous chemical elements at the rate 36 and 96 kg ha⁻¹, respectively. On sward pasture surfaces of fertilized and unfertilised treatment for preventing animal entrance, were built 15 iron boxes, fenced by 2.25 m² (1.5 m × 1.5 m) and 1 m high. The undisturbed area of sward inside the cages was used for assessing the herbages and soil characteristics reported further in the text, figures and tables. The dynamics of herbage development, flora composition and intake of herbage browsed, was determined by harvesting two samples of surface of 1 m² of the herbage present in side and 1 m² outside the iron box cage, at five phenological stages of development:

Fig. 1 Monthly mean (first letter of month in the graph) of meteorological characteristics (a, rainfall; b, mean temperature; c, evapotranspiration; and d, solar radiation) verified in the two years of evaluation.

herbaceous (November), beginning of shooting (March), heading (April), flowering (May) and dead plant (June). The harvest of each phonological stage was three times replicated. After harvest, the biomass inside and stubble after browsed outside the cage were weight and the areas inside the cage were cleaned from remnant herbage. From inside cages biomass samples of about 1,000 g were randomly kept, dried in an air-forced oven at 65 °C for 72 h for determining moisture content at harvest and afterwards ground with Cyclotec mill with a mesh screen with Ø of 1 mm. The milled samples were stored in room temperature until

laboratory analyses. At the end of experiment the nutritive values of flour were determined by chemically analyses: crude protein (CP, Kirsten [18]), crude fibre (CF, Henneberg and Stohmann [19]), neutral-detergent fibre (NDF), acid-detergent fibre (ADF) and acid-detergent lignin (ADL, Goering and van Soest [20]). The milk forage unit (MFU) values of the herbage were assessed according to the method used by Andrieu and Weiss [21].

The second samples of 1 m⁻² biomass harvest, were used to determine flora composition. The biomass of the each sample was botanically sieved and the flora

classified according to Fiori [22] and Pignatti [23] botanical guide. The flora species were grouped into Graminaceae, Leguminosae and Compositae botanical families and a group of miscellaneous, which included species of the Boraginaceae, Ranunculaceae, Scrophulariaceae and Umbelliferae families, whose proportion in the DM was less than 3%. The separated samples were weighed, dried and their contribution to DM herbage was determined.

Every 20 days, the animals at grazing and those in cattleshed were weighted and 30 mL of blood sample were collected from each buffalo heifers for determination of biochemical profile in order to evaluate the animal health and to control the puberty status.

The soil characteristics, before and at end the trails, were assessed on a profile of 0-30 cm Ap horizon. Before fertilizer application, in the side 4 cages of native and improved sward were harvested 4 core samples of soil before pasture fertilizing in 2005 and after the last harvest of 2007. The samples were air-dried and, after root separation, were sieved through a mesh screen of 2 mm of Ø, and later used for assessing pH (1:2.5 soil:water ratio) [24], total nitrogen [25], organic matter [26], phosphorous [27, 28], and potassium [29].

Statistical analysis adopted for processing the data of soil enrichment on dynamics of biomass development, flora composition and qualitative characteristics of phonological stage was ANOVA techniques according to a factorial design with three replications. The priority among factors of factorial model was: first, year (e.g. 2 levels); second, environments (e.g. 2 levels) third, fertilizer (e.g. 2 levels) and fourth, harvest at specific phonological stage of biomass development (e.g. 5 levels). The model considers environment and replication as fixed effects and year and fertilizer as random effect [30]. Because the interaction effects of year with factors (environment and fertilizer application) were not significant, the data presented in the results, tables and figures are shown as a mean of the years.

Comparisons among harvest of phonological stage in native and improved sward were made using Fisher's protected last significant difference (LSD) at $P \geq 0.05$ level of probability.

The effects of fertilizer application on biochemical characteristics of topsoil were determined processing the data recorded at the beginning and after experiment according to factorial design with three factors: year, environment and fertilizer with two levels and three replications. Comparison of biochemical compounds among environments and fertiliser treatment on sward were tested by Fisher's protected last significant difference (LSD) at $P \geq 0.05$ level of probability.

Statistical analyses of data related to weight body gain of the heifers bred in the pasture and those in cattleshed were conducted separately by using PROC ANOVA procedure of SAS [31]. The weight body gain was determined for each heifer by divided the difference of weight at the end and those of the beginning by number of days which animal browsed the sward of pastures. In the analysis, each heifer is considered a replication of the experiment. The weight body gain data were analysed according to factorial design with two factors: environment (e.g. 2 level) and animal feeding (e.g. 3 level). Comparison among the mean weight body gain of animals fed by browsing in the native and improved sward and unifeed in cattleshed, were tested with Duncun's Multiple Range Tests at $P \geq 0.05$ probability level.

3. Results

The mean over year of dry matter production of native pasture at flowering phonological stage of flora in the sward of E I was about 72.6% lower than those of E II (Fig. 2). The effect of agronomic treatment increased the forage yield in 2006 and 2007 by 36.9% and 40.6% in E I and 32.6% and 34.8% in E II, respectively (Fig. 2). The reduction in dry matter production observed the second year of evaluation (7.5% in E I and 3.4% in E II) was a consequences of

Fig. 2 Dry matter production of pastures in environments across the years of evaluation at flowering stage of grasses.

favourable weather condition development in the first year (rainfall, mean temperature, Eto and solar radiation) (Fig. 1) while the discrepancy observed in yielding of E I and E II pastures were ascribed to a favourable weather (higher rainfall, lower mean temperature, Eto and solar radiation) and biochemical topsoil characteristics (lower pH, higher organic carbon and C/N) observed in E II rather than E I (Fig. 1, Table 1).

The effect of agronomic treatment differently influenced the dynamic of flora development across the period of growing in E I and E II (Table 2). However, because the dynamics of biomass development under both improved pastures was, in each phenological stage of harvest, higher than those of native sward, the

agronomic treatment did not interfere with weather environmental condition occurred during vegetative herbage development. The biomass produced in the phenological stage of the I, II and V harvest (November, March and June) was higher browsed, in both environments and kind of sward, than those III and IV harvest (April-May) and the unbrowsed biomass (stubbed after browsing) was related to the herbage developed in the sward (Table 1).

The moisture at I harvest to III harvest of native and improved sward range from 79.1% to 87.7% and was not statistically significant (Fig. 3). The variation in moisture content among I to III harvests of E I and E II was a consequences of flora composition and development of biomass in the experiments. The

Table 1 Biochemical characteristics of topsoil horizon (Ap 0-30 cm) of the sward.

Site	pH	Organic carbon (g kg ⁻¹)	Total nitrogen (g kg ⁻¹)	Phosphorous (ppm kg ⁻¹)	Potassium (ppm kg ⁻¹)	C/N
Environment I						
Native pasture	8.2	14.6	2.20	15	860	8.2
Improved pasture	7.5	14.3	1.63	14	858	8.3
LSD 0.05	NS	NS	0.21	NS	NS	NS
Environment II						
Native pasture	7.4	16.8	2.17	14	1,786	9.9
Improved pasture	7.3	16.9	3.80	16	1,965	9.4
LSD 0.05	NS	NS	0.12	NS	NS	NS

Table 2 Dynamics of dry matter production and animal biomass browsed.

Harvest	E I dry matter (g m ⁻²)				E II dry matter (g m ⁻²)			
	Native sward		Improved sward		Native sward		Improved sward	
	Undisturbed	Stubbed	Undisturbed	Stubbed	Undisturbed	Stubbed	Undisturbed	Stubbed
I	177	--	135	--	337	--	616	--
II	234	31	518	99	556	285	1,045	353
III	425	250	675	361	1,535	353	2,278	559
IV	883	632	1,064	800	2,640	128	4,901	523
V	637	113	1,098	161	1,284	90	1,861	342
LSD 0.05	26	47	27	43	39	42	43	56

Mean between natural and improved sward in the environment with the same letter are not statistical significant at Duncan's Multiple Range Test at P=0.05 level of probability. NS not significant.

Fig. 3 Mean moisture over the years of biomass at harvests in 5 phenological stages of herbage development (I Harvest, herbaceous; II Harvest, beginning of shooting; III Harvest, heading; IV Harvest, flowering; V Harvest, dead plant).

decrease of moisture mean percentage from about 66.5% to 50.0% in the IV and V harvest was due to the reduced physiological activity of the biomass with the increase of weather parameters (Figs. 1b-d and 3). Furthermore, the higher decrease of moisture as effect of weather aridity in the IV harvest of improved pasture and V harvests of improved in E II and V

harvest in E I rather than native herbage, was a consequences of higher herbage production of improved pasture which stressed the water available in topsoil for satisfying the physiological process than those of native pasture [32]. The higher herbage production of improved pasture stressed the water available in topsoil for satisfying the physiological

process than those of native pasture [7]. Thus, as consequences of higher amount of water requirement for respiration process the herbage of improved pastures dead before those of the native sward. However, the aridity condition favoured by the increase of weather parameters temperature, Eto and solar radiation and the reduced rainfall in the months June to August in E I and E II, affect the vegetative development of flora herbage reducing the physiological process of plant growth [27, 32]. The recovering of condition of plant growing, occurred with water felt in September in E II and October in E I 33 and 72 mm, respectively (Fig. 1a) and reduction of mean temperature, Eto and solar radiation (Fig. 1 b, c, d), breaking up the dormancy of herbage favouring plant growth whose dry matter in the I harvest in E II was 47.5% in native and 59.9% in improved pasture higher than E I (Table 2).

The pasture under study differed by flora composition of the sward and the amount of species was related to period of grass development (Table 3). The fertilizer enrichment in E I increased the mean over the harvests by 39.6% of Graminaceae and reduced by 41.1% and 30% the content of Leguminosae and miscellaneous group, respectively.

The flora of Graminaceae prevailed in both native pastures of the E I and E II; the diffusion of the flora of these botanical family was reduced passing from I to IV harvests in E I and in E II. However, the percentage of Graminaceae species in the I and II harvests in native and improved sward of E I and E II

were higher than the species of other botanical families. The amount of percentage of Leguminosae in native and improved sward of E I was 35.1% and 78.5% higher than those of E II while the mean of percentage of species belonged to Compositae across the harvests of the native sward was 77.1% lower in E I and 51.0% higher in E II than miscellaneous. By contrast, under improved sward the percentage of Compositae vs miscellaneous was 62.2% and 83.0% in E I and EII, respectively (Table 3). The mean of Compositae species over the harvest of native pasture in E II was 23% higher and 14% lower in the miscellaneous group than those of the native pasture of E I (Table 3).

The Graminaceae species were higher represented in both sward of E I and E II in the I and II harvest (mean 58% and 69.5% in E I; and 55.5% and 66.0% in E II in native and improved sward, respectively) rather than the following harvests while the species of Leguminosae, Compositae and miscellaneous group, in native and improved sward of E I and E II, were more represented in the III harvest (Table 3).

The variation of flora composition achieved by soil enrichment influenced the trait CP, CF, ADL and MFU of the herbage [33]. In the harvests of E I and E II, the content of CP and CF were related to age of plant development. The content of native sward of E I was 2.7% higher and 8.8% lower than the values of E II, respectively (Table 4). The average over harvests of CP of improved sward was increased by 13.8% in E I and 13.1% in E II (Table 4). The reduction of CP

Table 3 Evolution of flora composition across the dynamics development of the herbage.

Harvest	E I flora composition (%)								E II flora composition (%)							
	Native sward				Improved sward				Native sward				Improved sward			
	G*%	L*%	C*%	M*%	G*%	L*%	C*%	M*%	G*%	L*%	C*%	M*%	G*%	L*%	C*%	M*%
I	65	4	1	30	69	7	18	7	63	4	23	10	89	2	2	7
II	51	24	1	25	70	11	3	16	48	1	36	15	43	3	3	51
III	13	41	5	41	30	22	7	41	40	20	24	15	72	4	3	21
IV	34	37	11	8	52	18	1	28	31	15	44	10	89	3	7	1
V	27	23	14	36	74	12	8	6	43	18	18	21	76	2	2	20
LSD 0.05	6	8	3	10	8	5	7	9	8	7	4	3	10	1	1	4

* Botanical family: G = Graminaceae; L = Leguminosae; C = Compositae; M = Miscellaneous species.

Table 4 Main qualitative components of the herbage across the dynamic plant development.

Harvest	E I qualitative traits of biomass								E II qualitative traits of biomass							
	Native sward*				Improved sward*				Native sward*				Improved sward*			
	CP%	CF%	ADL%	MFU n	CP%	CF%	ADL%	MFU n	CP%	CF%	ADL%	MFU n	CP%	CF%	ADL%	MFU n
I	21	18	12	0.52	19	17	6	0.62	18	18	6	0.60	24	24	5	0.57
II	13	19	6	0.58	28	21	7	0.66	21	26	6	0.58	24	26	6	0.59
III	17	19	8	0.52	19	23	8	0.46	17	26	9	0.54	16	24	7	0.50
IV	14	22	7	0.52	13	25	9	0.56	11	23	11	0.37	10	28	10	0.43
V	10	26	7	0.49	8	19	8	0.41	6	21	9	0.37	10	21	8	0.45
LSD 0.05	4	3	2	0.02	5	2	2	0.01	4	2	2	0.01	2	3	2	0.01

* Qualitative component: CP = Crude protein; CF = Crude fibre; ADL = Acid detergent lignin; MFU = Milk forage unit.

through the harvest, in native and improved sward, showed the same trend evidencing that chemical application play a similar role on physiological process of herbage growing while the decrease of MUF was a consequence of increase of structural organ of plant development (CF and ADL) (Table 4). However on average of harvests, the MFU of native sward was lower than 3.0% in E I and 3.2% in E II than improved sward (Table 4). Thus, as consequences of higher MFU availability per unit area of improved than native sward (MFU m⁻² mean over harvests: 248 and 625 in native and 378 and 1,087 in improve sward of E I and E II, respectively) (Tables 2, 4).

The effect of higher MFU m⁻² availability of improved than those of native sward increased the weight body gain of heifers (Fig. 4).

The weight of body gain per day at the end of experiment of the heifers which had browsed the sward of native and improved pasture was higher 27.6% in native and 26.8% in proved sward of E II rather than E I, respectively (Fig. 4). The weight body gain of animal at grazing in comparison to those bred in cattleshed was lower 40% and 19.4% in native and 25.7% and 1.4% in improved of E I and E II pastures, respectively than the buffalo heifers bred in cattleshed (Fig. 4).

4. Discussion

The adopted chemical soil enrichment influenced the pastures herbage favouring an increase of biomass and nutritive value of protected EU Mediterranean pasturelands [8, 13, 34]. The low variation of topsoil

Mean with the same letter are not statistical significant at Duncan's Multiple Range Test at P=0.05 level of probability.

Fig. 4 Mean of weight body gain (g day⁻¹) of buffalo heifers browsed on native and improved sward in compared with animal bred in cattleshed and fed with unified.

biochemical parameters of enriched sward at end of the experiments evidenced that the elements allowed with agronomic improvement sustaining the increase of dry matter and feeding characteristics of the improved sward (Table 1) [12, 13, 34]. According to the results obtained in other environments by Norman [9], Osman et al. [34], Sneva et al. [35] and Tupper [36] the topsoil enriched with nitrogen and phosphorous favoured higher DM yield and MFU than the native pasture (mean of enriched sward of E I and II was 56.5% and 23.2% higher than native sward, respectively) (Fig. 2, Table 2). The low variation of soil organic carbon at the end of the experiment between native and improved treatment as reported by Selling et al. [37], Sanchez-Maran et al. [38], Rutigliano et al. [39], and Li et al. [40] may evidence an increase of soil microbial activity which favour a release of nutrients cycling for sustaining the increase of biomass production across the dynamic phenological phase (Fig. 2, Table 2). The induced changes in the ratio among botanical phytocoenoses of the native pasture observed in improved sward may be due to the effect of enriched soil fertility on seedbank (Table 3). Experimental studies carried out in British Columbia (Canada) [10], south-eastern Australia [7, 13, 36], western Oklahoma (USA) [41], Argentina [42] and Italy [33] showed that the soil fertilizer application differently influenced the residual effect, dry matter production, quality of herbage and flora composition of the sward.

However, in literature it is reported that the effect of topsoil enrichment may favour recover of soil seed-bank born from dormant vegetative stasis in topsoil whose amount has been estimated, in some pasture of USA, to be roughly more than 44% of the flora present in the pasturelands [43, 44].

According to Pérez et al. [45], Nie et al. [46], Tracy and Sanderson [43] and Iannucci et al. [47], the higher increase of dry matter in improved sward of E I and E II may be a consequence of the effect of fertilizers soil enrichment which favoured development of native

sward and recover of seedbank flora of Compositae, Graminaceae, Leguminosae and miscellaneous botanical family. Thus, the effect of soil enrichment on seed-bank may represent the agronomic treatment able to satisfy the expectation of the EU directive [17].

The agronomic approach adopted in the study favoured the development of Leguminosae rather than flora of others families with a consequent increase of CP content in the herbage of improved sward (13.8% and 13.1% in E I and E II, respectively) (Table 4). However, the higher concentration of NDF and ADF (data not shown) observed in the herbage under chemical enrichments was a consequence of greater morphological structure than native pasture (data not shown, mean E I and E II plant height of improved sward 38.8% than native sward).

According to McKenzie et al. [7], Johnston et al. [14] and Jouven et al. [48] the lower content of ADL (3.0% than improved sward) of native sward of E I and E II was due to the reduced percentage of Graminaceae (native sward lower 36.6% than improved) in the biomass of improved pasture which influence the quality of structural fibre of the herbage (Table 4). The higher gain in MFU (mean over the harvest) of improved than the native sward (3.0%, and 3.2% MFU in E I and E II, respectively) was a consequence of chemical enrichment of pastureland which induce variation in biomass yield and increase of qualitative characteristics of the browsed herbage (CP, CF, NDF, ADF and ADL) favouring a higher production of MFU of improved pasture.

Furthermore, the effect of heifers browsing of native and improved sward compared with those bred in cattleshed affect welfare, health, physiological puberty and morphological body properties [49-51].

Experimental results, on data concerning the effect of heifers bred browsing herbage of natural or improved sward compared to the animal raised up in cattleshed evidenced favourable change in the body of young heifers whose advantages on welfare has been reported by Napolitano et al. [49] and those on body

performance and health and animal fertility and puberty by Terzano et al. [50, 51].

5. Conclusions

The effect of improved pastures of animal grazing on increase of daily weight body gain was related to the herbage potential of native pasture. However, grazing heifers when compared with those bred in cattleshed showed improved welfare, body performance and health and animal fertility of the young buffaloes. Considering the effect of nitrogen and phosphorous enrichment on herbage and biodiversity of the flora in the sward and the effect of grazing animal on body condition of young heifer at first birth, the agronomic practices adopted in the study may be considered as a useful approach to increase the biomass and qualitative characteristics of the herbage preserving biodiversity and landscape characteristics in the protected Mediterranean pasturelands of the EU environments.

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