

# LEGUMES

## Quantifying Morphological Stage to Predict the Nutritive Value in Sulla (*Hedysarum coronarium* L.)

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### ABSTRACT

A precise prediction of quality of legumes in the field would enable harvests to be conducted at appropriate nutritive composition levels. Our objective was to predict the nutritive value of sulla (*Hedysarum coronarium* L.) with the codified morphological stage and weather parameters at two Mediterranean sites (Ancona and Sassari, Italy) in 1996, 1997, and 1998. Herbage samples were collected at progressive morphological stages, from vegetative stage to seed setting. Mean stage by weight (MSW), dry matter yield (DMY), leaf/stem ratio (L/S), crude protein concentration (CP), neutral detergent fiber (NDF), gross energy (GE), and organic matter digestibility (OMD) were determined. Forage characteristics were regressed on growing degree days (GDD) and MSW. The DMY ranged from 2 to more than 10 Mg ha<sup>-1</sup> from vegetative to seed set while L/S decreased from almost 5.6 to 0.2 and the CP from a maximum of 295 to a minimum of 107 g kg<sup>-1</sup> dry matter (DM). The NDF ranged from 200 to 616 g kg<sup>-1</sup> DM and was best predicted by L/S and MSW. The GE was relatively constant across growth stages with a mean value of 18.0 MJ kg<sup>-1</sup> DM. The OMD ranged from 398 to 846 g kg<sup>-1</sup> organic matter (OM) and declined linearly with increasing MSW. The OMD decreased 32.6 g kg<sup>-1</sup> OM per stage unit and followed similar trends for the two sites, with a lower level at the warmer site (Sassari). The MSW was a better predictor of sulla OMD than GDD, with a higher R<sup>2</sup> (0.70 vs. 0.54) and a lower root mean square error.

SULLA IS A SHORT-LIVED, perennial, herbaceous legume that originates from the Mediterranean basin (Talamucci, 1998) where it is grown as a 2-yr crop without irrigation. It is well adapted to semiarid environments in clay and calcareous soils. Sulla is extensively grown as a fodder in central and southern Italy (latitudes from 36° to 45° N) where it occupies around 150 000 ha (Talamucci, 1998). It is used as forage crop in other countries in the Mediterranean basin, such as Spain, Portugal, and Tunisia (Más, 1986; Trifi-Farah et al., 1989). Sulla

was introduced into New Zealand in 1949 for soil protection purposes (Watson, 1982) because of its favorable characteristics and is considered a suitable forage plant in New Zealand and Australia (Stienezen et al., 1996). Sulla is a high-yielding crop, up to 14 Mg ha<sup>-1</sup> DM per season (Stringi et al., 1997; Douglas et al., 1999), that produces forage mainly during early spring and autumn and is useful for grazing, haying, and ensiling (Sulas et al., 1995; Valente et al., 1999). It is a good source of protein for livestock and has a moderate extractable condensed tannin concentration that ranges from 25 to 40 g kg<sup>-1</sup> DM (Terrill et al., 1992; Douglas et al., 1999; Piluzza et al., 2000).

A better knowledge of the developmental morphology and quality changes of sulla in different environments is necessary to optimize its potential for livestock production. The variation in nutritive value of the herbage is related to the environmental and physiological history of the crop (Marten et al., 1988; Nelson and Moser, 1994). It is well known that forage maturity at the time of harvest is the primary factor influencing nutritive value within a species. Additionally, nutritive value has been related to accumulated GDD, crop age in days, L/S, and stand height. However, the codified stage of development was the most reliable in predicting the nutritive value over a wide range of crop and environmental conditions (Kalu and Fick, 1983; Fick and Onstad, 1988; Sanderson, 1992; Valente et al., 2000).

One method to quantify the morphological stage of alfalfa (*Medicago sativa* L.) is that of Kalu and Fick (1981), which proposes a numerical scale for individual stems and two alternative procedures to determine population mean stage based on shoot numbers (mean stage by count) and shoot weight (MSW). The codified morphological stage could be useful to predict herbage quality, determine the best time to cut or graze to attain a certain yield level or nutritive value, or both (Simon and Park, 1983). Furthermore, simulation models of forage production and systems analysis of forage management have included the prediction of forage quality as an essential ingredient in the process (Fick, 1984; Fick et al., 1994).

Although sulla is a productive crop with potentially high nutritive value for semiarid environments, information regarding the nutritive value as a function of

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**Abbreviations:** CP, crude protein concentration; DM, dry matter; DMY, dry matter yield; GDD, growing degree day(s); GE, gross energy; JDAY, day of the year; L/S, leaf/stem ratio; MSW, mean stage by weight; NDF, neutral detergent fiber; OM, organic matter; OMD, organic matter digestibility; RMSE, root mean square error; TLIT, total hours of light for growth from 1 January.

**Table 1. Environmental and soil characteristics of Ancona and Sassari, Italy.**

Property	Site	
	Ancona	Sassari
Latitude	43°32' N	40°53' N
Longitude	13°23' E	8°29' E
Elevation, m (above sea level)	203	60
Annual mean temperature, °C	13.7	16.2
Annual precipitation, mm	765	547
<b>Soil characteristics</b>		
Gravel, %	0	16
Sand, %	14	48
Silt, %	40	23
Clay, %	46	13
pH, 1:2.5 w/v soil:water	8.1	7.4
Total N, %	0.15	0.14
Organic C, %	0.86	1.2

weather and crop parameters is very limited. The objective of this study was to evaluate MSW, L/S, GDD, total hours of above-horizon sunlight for the growth (TLIT), day of the year (JDAY), and DMY as predictors of CP, NDF, GE, and OMD of sulla over a 3-yr period at two different Mediterranean sites.

## MATERIALS AND METHODS

### Sites and Crop Management

The trials were conducted during a 3-yr period (1996–1998) at two sites with differing climates and soil types (Table 1). Stands of sulla were sown at 25 kg ha<sup>-1</sup> dehulled viable seed. Two sulla cultivars with different earliness, ‘Grimaldi’ and ‘Sparacia’, were used at Ancona while at Sassari only Grimaldi was used. Because the specific rhizobia are absent at Sassari, sulla was inoculated with a Sardinia strain of *Rhizobium hedy-sari* (Di.SAABA, University of Sassari, Italy). The two cultivars at Ancona were chosen because of their differences in earliness, Sparacia is earlier than Grimaldi. The seeding time and age of the sampled crops over the years are reported in Table 2. The stands sampled in 1996 and 1998 were second-year crops while the 1997 stand was a first-year crop. All plots were fertilized with 60 and 120 kg ha<sup>-1</sup> P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O, respectively, as recommended by soil test results. No irrigation, fertilizers, or herbicides were applied after sowing at either site. Temperature and rainfall data were collected from weather stations located 60 m from the Ancona experiment site and 100 m from the Sassari site, respectively.

Herbage samples were collected at progressive morphological stages from early vegetative to seed ripening, within one 0.5-m<sup>2</sup> quadrat randomly located in plots (8.0 by 8.0 m) cut to a standard height of 5 cm above ground level. Dry matter yield was determined on a 2-m<sup>2</sup> area inside the plots and dried to constant weight at 90°C in forced-air oven. Sampling was conducted at 1-wk intervals from March to the middle of June. The sampling started at Sassari on 30 Jan. 1998 due to favorable growing conditions and good crop development. The experiment at each site comprised three randomized complete blocks, and treatments were the ages (days after planting) of

**Table 3. Definition of morphological stages of development for individual sulla stems (modified from Kalu and Fick, 1981; Borreani et al., 1999).**

Code	Stage name	Stage definition
0	Rosette	No stems or floral buds
1	Midvegetative	Stem length <15 cm, no branches
2	Late vegetative	Stems >15 cm, no floral buds
3	Early bud	Appearance of first floral bud
4	Late bud	Floral buds, no open florets
5	Early flowering	First open floret
6	Flowering	Open florets, no green loment
7	Early seed set	Brown florets, first green loment
8	Seed set	Brown florets, green loment
9	Seed ripening	Ripe brown loment

the herbage. Vegetation on the plots to be harvested in 1998 was cut twice, once in the middle of May and once in fall of 1997.

### Morphological Stage Evaluation and Phytomass Partitioning

The morphological stage was evaluated on a sample of 50 shoots clipped at ground level and classified according to the 10-stage classification system developed by Kalu and Fick (1981) and slightly modified for sulla in the 0 to 2 stages by Borreani et al. (1999) (Table 3). The MSW was used to calculate the stage of development for the sulla populations using the following equation:

$$MSW = \sum (S \times D)/W$$

where  $S$  = morphological stage number (0–9);  $D$  = dry weight (90°C to constant weight) of the shoots in stage  $S$ , and  $W$  = total dry weight of the shoots in all stages.

The composition of the harvested herbage was also determined on 50 stems, dividing the plants into leaflets, petioles, stems, and racemes. The L/S was calculated on the dried material as (leaflets + petioles)/stems. The subdivision of the plant fractions was not performed at Sassari in 1996.

### Chemical Analysis

The herbage samples were immediately dried in a forced-draft oven to constant weight at 65°C (Deinum and Maassen, 1994) weighed, ground in a Cyclotec mill (Tecator, Herndon, VA) to pass a 1 mm screen, and stored at –18°C for qualitative analyses. The dried samples were analyzed to determine total N by combustion (Macro-N, Foss Heraeus Analysensysteme, Hanau, Germany) and NDF as described by Robertson and Van Soest (1981). Crude protein was determined by multiplying the N concentration of the samples by 6.25. Samples were also analyzed for GE with an adiabatic calorimeter bomb (IKA C7000, IKA, Staufen, Germany) and OMD, according to the two-stage rumen fluid technique (Tilley and Terry, 1963). The OMD values were expressed in vivo using the regression equation of Goldman et al. (1987).

### Statistical Analysis

The CP, NDF, GE, and OMD data were analyzed according to years, cultivars, harvest maturity, and sites by regression

**Table 2. Cultivar, seeding period, and age of sulla stand in 1996, 1997, and 1998 at Ancona and Sassari, Italy.**

Site	Cultivar	Seeding period	Stand Age		
			1996	1997	1998
Ancona	Grimaldi and Sparacia	Spring	Second year	–	–
Ancona	Grimaldi and Sparacia	Autumn	–	First year	Second year
Sassari	Grimaldi	Autumn	Second year	First year	Second year

**Table 4. Monthly and annual mean temperatures and accumulated precipitation for 1996, 1997, and 1998 and the 25-yr mean for Ancona and Sassari, Italy.**

Month	Ancona				Sassari			
	1996	1997	1998	25-yr mean†	1996	1997	1998	25-yr mean†
	Mean temperature, °C							
January	6.3	7.7	7.6	5.8	11.7	10.9	12.2	9.3
February	6.4	10.1	10.5	6.4	8.5	11.5	10.0	9.8
March	7.1	11.2	9.6	8.3	10	11.8	10.3	11.0
April	12.7	10.5	14.2	11.7	13.4	14.2	12.5	13.1
May	17.7	18.0	17.5	16.5	16.5	19.0	16.3	16.9
June	22.1	22.2	23.3	22.2	20.4	23.1	20.4	21.4
July	23.1	23.5	26.5	23.6	23.0	24.2	22.9	24.1
August	23.0	23.6	26.4	22.4	23.4	25.9	23.6	24.4
September	16.7	21.5	20.4	18.6	19.9	23.4	21.0	22.5
October	14.2	16.2	16.8	15.2	16.8	20.5	16.9	17.7
November	8.9	11.0	9.2	8.4	14.2	16.0	11.7	13.6
December	5.7	7.7	4.7	4.9	11.6	13.6	9.3	10.1
Annual	13.6	15.2	15.4	13.7	15.8	17.9	15.6	16.2
	Rainfall, mm							
January	20	23	50	55	46	48	26	49
February	56	84	55	56	60	6	18	50
March	50	32	39	64	65	9	41	55
April	53	113	80	65	34	23	106	52
May	37	19	62	56	70	45	26	32
June	16	58	39	60	53	22	25	14
July	21	29	15	43	2	0	0	5
August	209	53	45	60	5	24	7	10
September	56	27	53	68	65	8	70	46
October	93	65	135	73	53	54	99	76
November	125	124	147	91	79	102	63	88
December	45	41	118	85	77	33	50	70
Annual	781	668	838	777	609	374	531	547

† Mean on the period from 1974 to 1998.

analysis, utilizing the SPSS software package (Norusis, 1992). The data were averaged on the field replicates before statistical analysis; a data set of 131 values was therefore used to evaluate regression relationships. The data were regressed on the MSW, L/S, GDD with a 5°C base temperature computed from 1 January to sampling date, TLIT computed for the same time interval as GDD, JDAY, and DMY as independent variables. The accumulated GDD were calculated as the sum of the difference between the mean daily temperature and a base temperature of 5°C. Negative values were not included in the summation. Linear and quadratic regressions were compared using the Draper and Smith (1998) stepwise selection procedure to choose the best regression model at the 0.05 probability level. All of the determination coefficients ( $R^2$ ) reported in this paper were adjusted for degrees of freedom. The MANOVA analysis of covariance was used to verify the equivalence of the equations for the cultivars, years, and sites (Norusis, 1992). The regression lines related to each location were compared between years and cultivars and were pooled when not significantly different. The best equation was selected based on the coefficient of determination, root mean square error (RMSE), and Mallows'  $C_p$  statistic (Draper and Smith, 1998).

## RESULTS AND DISCUSSION

### Weather Data

The two sites differed in temperature and rainfall during the three experimental years (Table 4). The weather at Ancona for each year was similar to the 25-yr mean. At Sassari, 1996 and 1998 had spring and annual air temperatures and annual accumulated rain that were very similar to the 25-yr mean, whereas 1997 had lower rainfall and higher air temperatures.

The GDD at Sassari started to accumulate from January on, reaching around 400°C at the beginning of March. The GDD at Ancona started to accumulate 1 mo later than at Sassari and reached 400°C at the end of March, except for 1996 when lower GDD was observed in early spring compared with the other 2 yr. At the beginning of June, the GDD reached values of 781, 924, and 978°C at Ancona and 1072, 1301, and 1104°C at Sassari, for 1996, 1997, and 1998, respectively.

### Mean Stage by Weight, Leaf/Stem Ratio, and Dry Matter Yield

The MSW increased during the spring growing season with different trends in the 3 yr at both sites (Fig. 1). At Ancona, Grimaldi was earlier at the beginning of spring in 1998 than in 1996 (second-year crops, Table 2), because of the higher spring temperatures in 1998, while it reached flowering (MSW = 6) around the same date. The first-year crop (1997) reached flowering about 15 d later than the other 2 yr. Compared to Grimaldi, Sparacia showed similar trends and reached the same stages about 8 d earlier for the whole growth cycle in each year. At Sassari, Grimaldi reached flowering about 15 d earlier than at Ancona in 1998 while in 1996, it reached flowering at the same date as at Ancona because of low temperatures. The MSW increased with increasing GDD (Fig. 2). Both cultivars showed the same trends at Ancona in 1996 and 1998 while in 1997, stages were delayed. Grimaldi at Sassari showed a similar stage development in 1996 and 1997 while in 1998, it matured earlier due to a temperate winter that permitted uninterrupted growth.

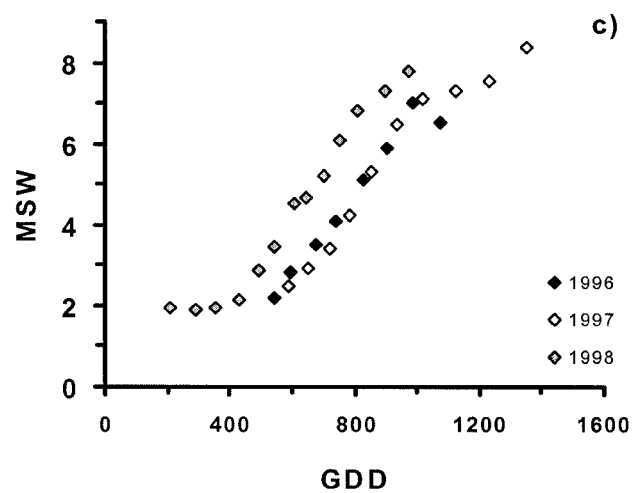
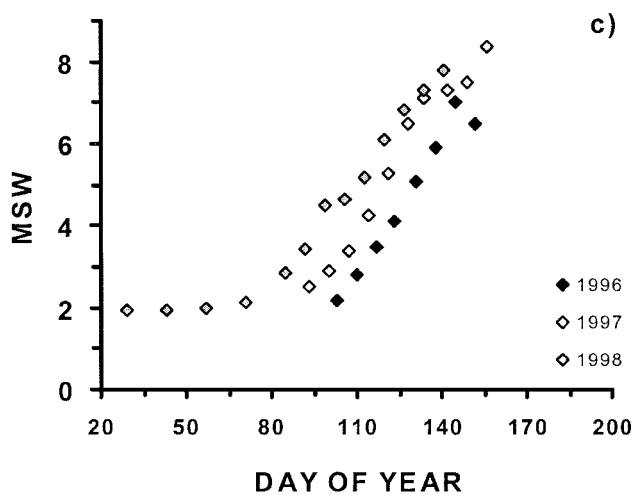
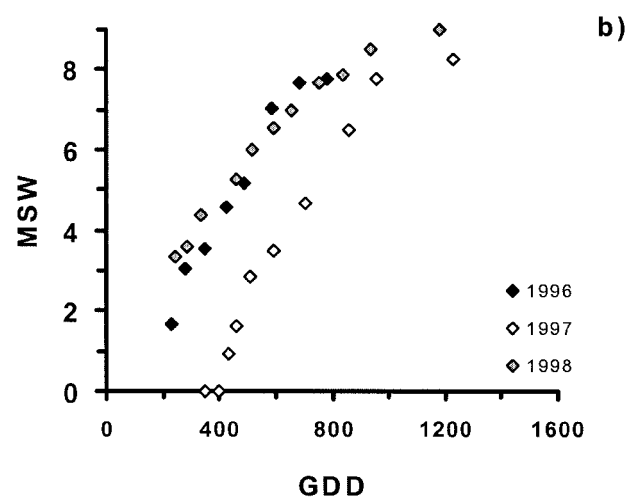
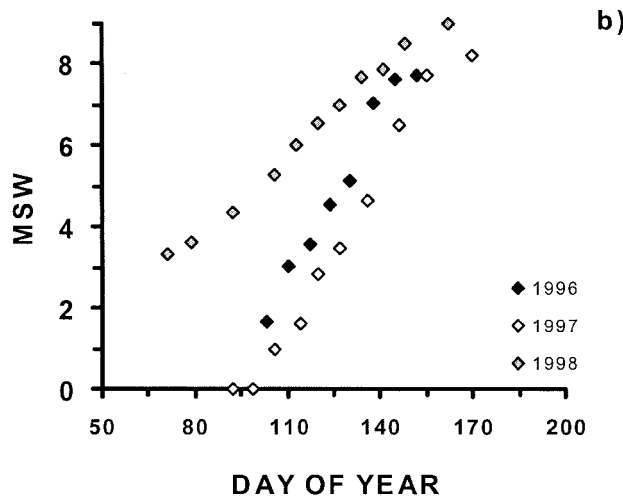
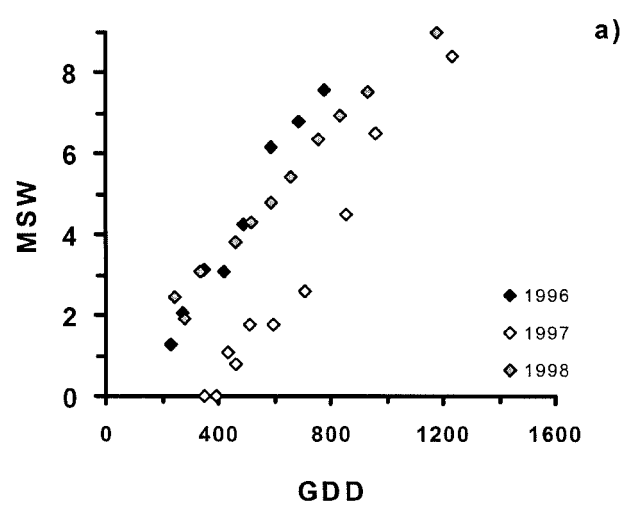
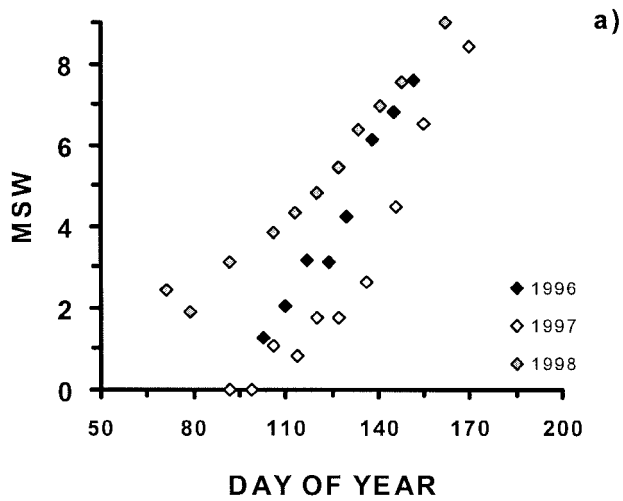


Fig. 1. Changes in sulla mean stage by weight (MSW) in relation to day of the year at two sites over 3 yr: (a) Ancona 'Grimaldi', (b) Ancona 'Sparacia', and (c) Sassari Grimaldi.

Fig. 2. Changes in sulla mean stage by weight (MSW) in relation to growing degree days (GDD) at two sites over 3 yr: (a) Ancona 'Grimaldi', (b) Ancona 'Sparacia', and (c) Sassari Grimaldi.

The L/S are reported in Fig. 3, starting from MSW = 2 because the ratio tends to infinite in less mature herbage. The L/S at Ancona decreased from 5.6 to 0.2 for Grimaldi, with similar trends in 1996 and 1998, but was

slightly higher in 1997. The L/S of Sparacia ranged from 0.3 to 5.1, with the same exponential trends over the years. Grimaldi L/S at Sassari was higher in 1997 (dry, warm) than in 1998, with values that were lower than

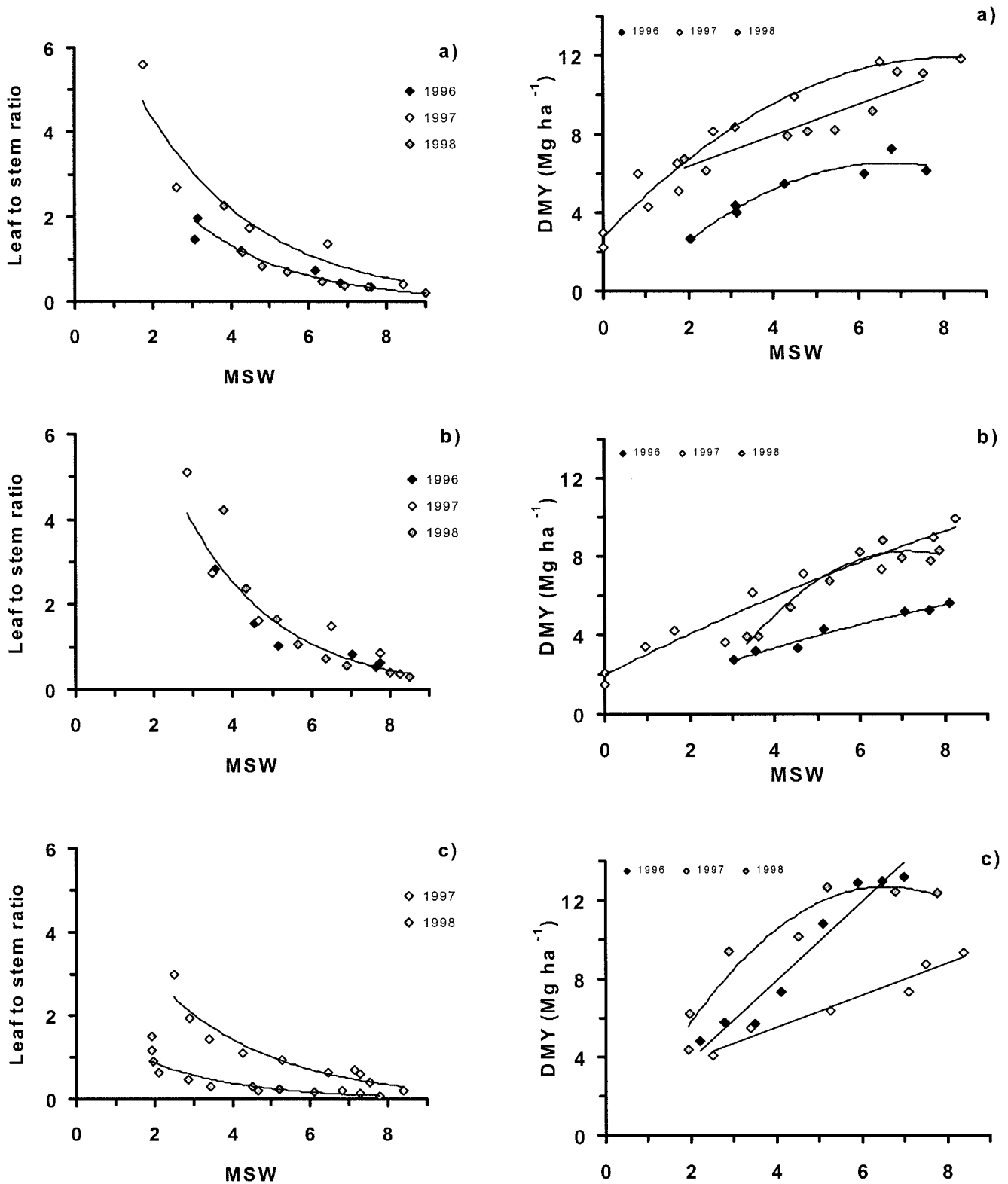


Fig. 3. Sulla leaf to stem ratio as a function of mean stage by weight (MSW) at two sites over 3 yr: (a) Ancona 'Grimaldi', (b) Ancona 'Sparacia', and (c) Sassari Grimaldi. Sampling was not performed at Sassari in 1996.

Fig. 4. Sulla dry matter yield (DMY) in relation to mean stage by weight (MSW) at two sites over 3 yr: (a) Ancona 'Grimaldi', (b) Ancona 'Sparacia', and (c) Sassari Grimaldi.

at Ancona at the same stage of growth. Higher L/S due to drought has been reported for other legumes and grasses (e.g., Vough and Marten, 1971; Lemaire et al., 1989).

The DMY of sulla increased rapidly as phenology advanced (Fig. 4). Grimaldi at Ancona produced more than Sparacia ( $P < 0.01$ ) when compared at the same MSW. Both cultivars resulted in a lower production in 1996 than in 1997 or 1998. Grimaldi at Sassari reached 12 Mg ha<sup>-1</sup> DM at flowering in 1996 and 1998 while in 1997, it did not exceed 9 Mg ha<sup>-1</sup> DM.

### Crude Protein Concentration and Neutral Detergent Fiber

Crude protein concentration was negatively related to MSW and, at Ancona, ranged from 117 to 262 g kg<sup>-1</sup> DM for Grimaldi and from 116 to 295 g kg<sup>-1</sup> DM for

Sparacia (Fig. 5). Sparacia CP was higher than that of Grimaldi by about 25 g kg<sup>-1</sup> DM in 1996 and 35 g kg<sup>-1</sup> DM in 1998, over the whole growth cycle, while in 1997, there were no differences. When analyzing the CP as a function of the DMY, no difference was found between the cultivars, due to the N dilution in the DM shoot biomass, as observed elsewhere for alfalfa (Lemaire et al., 1985). The CP of Grimaldi at Sassari ranged from 107 to 219 g kg<sup>-1</sup> DM, with no differences over years, and had a lower concentration than at Ancona.

The regression equations for predicting sulla CP from GDD, JDAY, TLIT, MSW, L/S, and DMY had coefficients of determination never higher than 0.62 (Table 5). The best predictor was GDD followed by DMY with an  $r^2$  of 0.62 and 0.56 and an RMSE of calibration of 25 and 24 for the two variables, respectively. The MSW had a linear correlation with an  $r^2$  of 0.47, which was

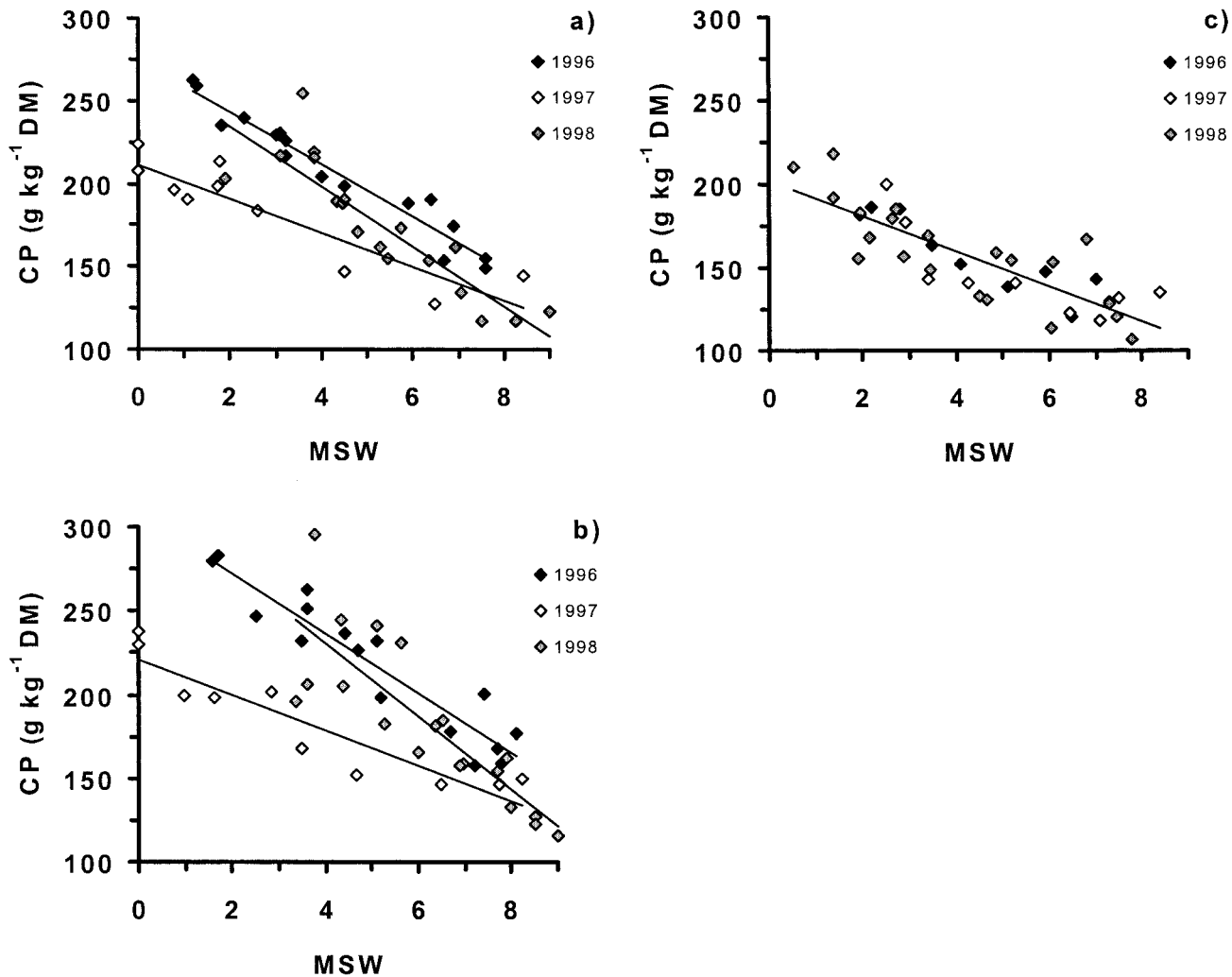


Fig. 5. Sulla crude protein concentration (CP) as a function of mean stage by weight (MSW) at two sites over 3 yr: (a) Ancona 'Grimaldi', (b) Ancona 'Sparacia', and (c) Sassari Grimaldi. Regression equations of the pooled data:

- (a)  $CP_{\text{Grimaldi } 96} \text{ (g kg}^{-1} \text{ DM)} = -15.7\text{MSW} + 274 \text{ } r^2 = 0.94 \text{ RMSE} = 9$   
 $CP_{\text{Grimaldi } 97} \text{ (g kg}^{-1} \text{ DM)} = -10.3\text{MSW} + 211 \text{ } r^2 = 0.80 \text{ RMSE} = 15$   
 $CP_{\text{Grimaldi } 98} \text{ (g kg}^{-1} \text{ DM)} = -18.0\text{MSW} + 271 \text{ } r^2 = 0.73 \text{ RMSE} = 20$   
 (b)  $CP_{\text{Sparacia } 96} \text{ (g kg}^{-1} \text{ DM)} = -17.9\text{MSW} + 308 \text{ } r^2 = 0.88 \text{ RMSE} = 14$   
 $CP_{\text{Sparacia } 97} \text{ (g kg}^{-1} \text{ DM)} = -10.5\text{MSW} + 221 \text{ } r^2 = 0.83 \text{ RMSE} = 14$   
 $CP_{\text{Sparacia } 98} \text{ (g kg}^{-1} \text{ DM)} = -21.8\text{MSW} + 318 \text{ } r^2 = 0.68 \text{ RMSE} = 26$   
 (c)  $CP_{\text{Sassari } 96-98} \text{ (g kg}^{-1} \text{ DM)} = -8.57\text{MSW} + 191 \text{ } r^2 = 0.57 \text{ RMSE} = 15$

**Table 5.** Regression equations, coefficients of determination, and root mean square errors (RMSE) for predicting crude protein (CP, g kg<sup>-1</sup> dry matter), neutral detergent fiber (NDF, g kg<sup>-1</sup> dry matter), and organic matter digestibility (OMD, g kg<sup>-1</sup> organic matter) with dry matter yield (DMY, Mg ha<sup>-1</sup>), growing degree days (GDD), day of the year (JDAY), leaf/stem ratio (L/S), mean stage by weight (MSW), and total hours of light for the growth from 1 January (TLIT) for sulla grown near Ancona and Sassari, Italy, in 1996, 1997, and 1998 ( $n = 131$ ).

Parameter	Equation model†	Adjusted $r^2$ or $R^2$	RMSE
CP	$-0.015 \text{ JDAY}^2 + 2.70 \text{ JDAY} + 87.5$	0.31**	34
CP	$-0.000085 \text{ TLIT}^2 + 0.167 \text{ TLIT} + 123$	0.31**	34
CP	$+162 \text{ L/S}^{0.0948}$	0.47**†	31
CP	$-11.6 \text{ MSW} + 235$	0.47**	30
CP	$+1.04 \text{ DMY}^2 - 24.2 \text{ DMY} + 282$	0.56**	24
CP	$+0.000078 \text{ GDD}^2 - 0.227 \text{ GDD} + 288$	0.62**	25
NDF	$+16.6 \text{ DMY} + 314$	0.30**	76
NDF	$+0.032 \text{ JDAY}^2 - 0.509 \text{ JDAY} + 545$	0.40**	71
NDF	$+0.00017 \text{ TLIT}^2 - 0.280 \text{ TLIT} + 462$	0.40**	71
NDF	$+0.231 \text{ GDD} + 264$	0.43**	69
NDF	$+28.4 \text{ MSW} + 284$	0.54**	62
NDF	$+420 \text{ L/S}^{-0.1465}$	0.67**†	54
OMD	$-2.33 \text{ JDAY} + 920$	0.37**	73
OMD	$-0.181 \text{ TLIT} + 887$	0.39**	72
OMD	$-21.6 \text{ DMY} + 779$	0.42**	73
OMD	$-0.261 \text{ GDD} + 805$	0.54**	63
OMD	$+612 \text{ L/S}^{0.0927}$	0.61**†	61
OMD	$-32.6 \text{ MSW} + 784$	0.70**	51

\*\* Significant at  $P < 0.01$ .

† Reduced data set ( $n = 69$ ).

lower than that obtained with GDD and DMY. The L/S, for which a complete data set is not available, had an exponential relationship with an  $r^2$  of 0.47 and an RMSE of 31.

Although sulla is a N-fixing legume and did not receive N fertilization, CP varied for the cultivars over the years and between the sites. This is consistent with the influence of many environmental and plant factors on CP content, as observed in alfalfa by Fick and Onstad (1988) and Sanderson (1992). This variability makes it difficult to identify a single parameter that is useful for predicting the CP content of forage legumes, as reported by other researchers working in this field (Onstad and Fick, 1983; Owens et al., 1995).

Throughout the growing season, the NDF at Ancona ranged from 200 to 616 g kg<sup>-1</sup> DM for Grimaldi and from 234 to 576 for Sparacia (Fig. 6) while at Sassari, it ranged from 223 to 588. The regression equations for predicting sulla NDF from GDD, JDAY, TLIT, MSW, L/S, and DMY had coefficients of determination from 0.30 to 0.67 (Table 5). The NDF was best predicted by an exponential L/S model, which accounted for 70% of the variability in the NDF, whereas a linear MSW model accounted for 54% of the variability in the NDF.

### Gross Energy and Organic Matter Digestibility

Herbage GE remained constant during the growing cycle, with a mean value of  $18.0 \pm 0.25$  MJ kg<sup>-1</sup> DM and no differences among cultivars, years, or sites. This is consistent with other studies on alfalfa (Borreani et al., 2000) and Italian ryegrass (*Lolium multiflorum* Lam.) (Valente et al., 2000). Since GE was nearly constant across growth stages, OMD may be considered the most important forage factor determining energy value (Hvelplund et al., 1995). Therefore, the accurate prediction of OMD of the crop in the field using an easy and reliable parameter gives an indication of the nutritive value of the forage.

The OMD of the herbage was highly variable at both sites, varying from 398 to 846 g kg<sup>-1</sup> OM at Ancona and from 449 to 821 g kg<sup>-1</sup> OM at Sassari (Fig. 7). Because the two cultivars at Ancona did not differ when compared at the same growth stage, the equations reported in Fig. 5a have been calculated from the pooled data for each year of the study. The OMD was inversely related to MSW, with a high coefficient of determination and a low RMSE each year at both sites. The 1996 and 1997 crops at Ancona had similar OMD trends while in 1998, a faster OMD decline was observed than for the other 2 yr. The higher temperatures in April 1998 at Ancona (Table 4) may have reduced the digestibility pattern, as observed in other forage legumes such as alfalfa (Marten et al., 1988). The 1996 and 1998 crops at Sassari showed similar trends while the first-year crop, grown in a droughty year (1997), had a higher OMD of about 100 g kg<sup>-1</sup> OM (Fig. 7b). This increased OMD may be induced by drought stress (Van Soest, 1994). In many cases, water stress improves digestibility because of both an increased L/S (Fig. 3) and a higher digestibility of leaf and stem fractions (Nelson and Moser, 1994).

Trends with equal slopes and different intercepts were obtained when the OMD data of the two sites were compared ( $\text{OMD} = -33.8\text{MSW} + 806$ ;  $r^2 = 0.83$  for Ancona vs.  $\text{OMD} = -30.3\text{MSW} + 746$ ;  $r^2 = 0.58$  for Sassari). The warmer site (Sassari) had a lower mean OMD of about 29.5 g kg<sup>-1</sup> OM. The differences between the two sites could be due to differences in temperature (annual mean temperature 13.7°C vs. 16.2°C for Ancona and Sassari, respectively). In this experiment, a mean decrease in OMD of 11.8 g kg<sup>-1</sup> OM for each increase in 1°C was observed. In temperate and tropical forage grasses, OMD decreases from 5.0 to 11.4 g kg<sup>-1</sup> OM for each degree Celsius increase in temperature (Deinum et al., 1968; Minson and McLeod, 1970). The steep decrease in digestibility observed for sulla in advanced stages of maturity was higher than that of temperate

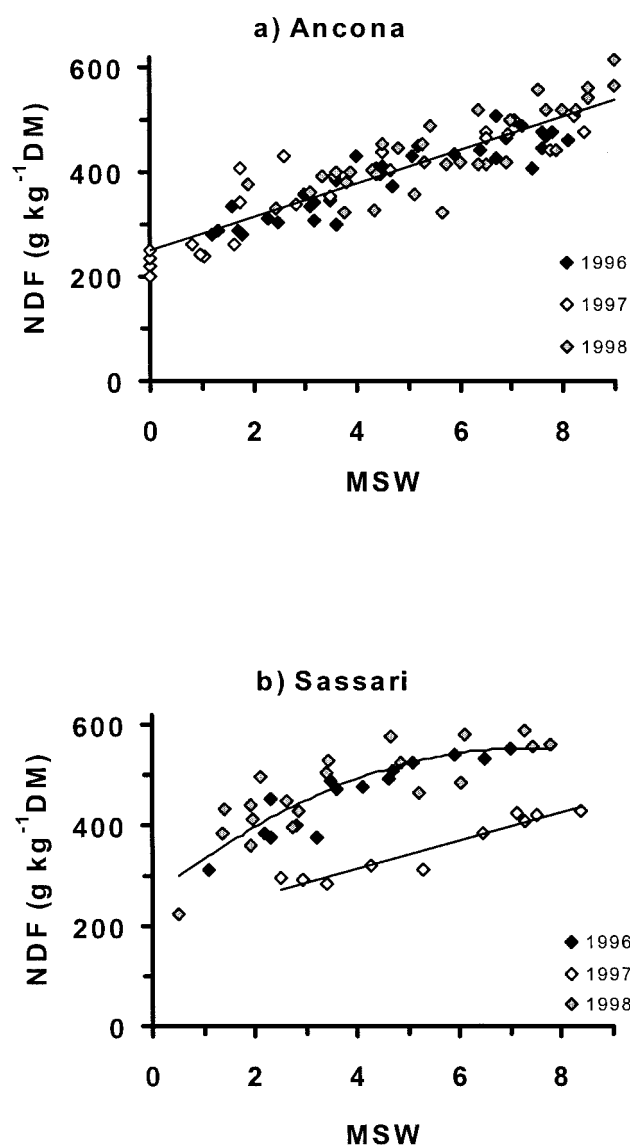


Fig. 6. Sulla neutral detergent fiber (NDF) as a function of mean stage by weight (MSW) at two sites over 3 yr: (a) Ancona (both cultivars) and (b) Sassari. Regression equations of the pooled data:

- (a)  $NDF_{Ancona} (g\ kg^{-1}\ DM) = 31.9MSW + 252\ r^2 = 0.83\ RMSE = 38$   
 (b)  $NDF_{Sassari\ 96-98} (g\ kg^{-1}\ DM) = -5.43MSW^2 + 80.1MSW + 258$   
 $r^2 = 0.73\ RMSE = 43$   
 $NDF_{Sassari\ 97} (g\ kg^{-1}\ DM) = 27.3MSW + 206\ r^2 = 0.91$   
 $RMSE = 19$

legumes such as alfalfa (Marten et al., 1988), which has contributed to the decline of sulla in the Mediterranean basin where it is traditionally harvested as hay at later stages than early seed set ( $MSW > 7$ ) with a preharvest OMD often lower than  $400\ g\ kg^{-1}\ OM$ . Only an efficient conservation technique such as ensiling would allow sulla to be harvested at earlier stages with high OMD and CP (Valente et al., 1999).

The regression equations for predicting sulla OMD from GDD, JDAY, TLIT, MSW, L/S, and DMY had coefficients of determination from 0.37 to 0.70 (Table 5). The best predictor ( $P < 0.05$ ) between the weather and crop parameters was MSW followed by L/S and GDD. The difference in prediction between MSW and GDD

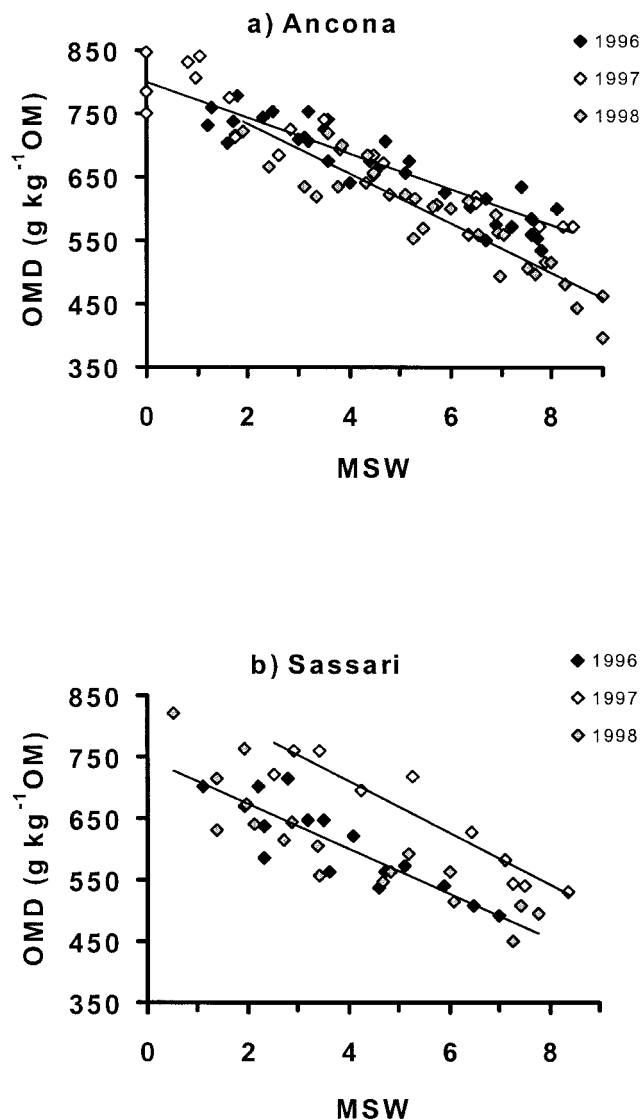


Fig. 7. Sulla organic matter digestibility (OMD) as a function of mean stage by weight (MSW) at two sites over 3 yr: (a) Ancona (both cultivars) and (b) Sassari. Regression equations of the pooled data:

- (a)  $OMD_{Ancona\ 96-97} (g\ kg^{-1}\ OM) = -29.3MSW + 802\ r^2 = 0.86$   
 $RMSE = 31$   
 $OMD_{Ancona\ 98} (g\ kg^{-1}\ OM) = -38.9MSW + 811\ r^2 = 0.82$   
 $RMSE = 34$   
 (b)  $OMD_{Sassari\ 96-98} (g\ kg^{-1}\ OM) = -36.7MSW + 746\ r^2 = 0.76$   
 $RMSE = 39$   
 $OMD_{Sassari\ 97} (g\ kg^{-1}\ OM) = -41.4MSW + 876\ r^2 = 0.88$   
 $RMSE = 33$

( $R^2$  of 0.70 vs. 0.54; RMSE of 51 vs. 63, respectively) was partly due to the different maturity of the two cultivars grown at Ancona. Comparing cultivars at the same growth stage is advantageous because it prevents differences in quality due to different L/S resulting from maturity from being attributed to the genotype. The L/S is less useful than MSW in predicting OMD and is a much more time-consuming method to predict quality directly in the field. Dry matter yield, which was first proposed in 1984 (Lemaire and Salette, 1984; Lemaire et al., 1985) to explain N content of grass and legumes, was not a good predictor of OMD. Mean stage by weight and

GDD were better in predicting OMD than CP. This is consistent with the compensatory effects on OMD of the individual chemical components of the herbage (protein, carbohydrates, and fiber fractions), which are individually influenced by environmental parameters (Van Soest, 1994).

## CONCLUSIONS

Sulla had high nutritive value in terms of CP and OMD until the late bud stage (MSW = 4), with DMY exceeding 4 Mg ha<sup>-1</sup> yr<sup>-1</sup>. Morphological stage was the most important factor in determining changes in OMD of herbage in the field, yet it was unable to accurately predict CP. The best predictor of CP was GDD even though there was only a moderate coefficient of determination. This indicates how difficult it is to find a single parameter to accurately predict the CP of sulla.

Mean stage by weight was a good predictor of the nutritive value and can be easily estimated by the grower. Furthermore, MSW appears to be a promising tool for rapid evaluation of forage quality, which is needed to properly manage the crop in semiarid environments. However, the decline in digestibility observed at the warmer site indicates that equations based on MSW to predict nutritive value should be developed for specific environments. If validated with other data sets, the equations based on MSW could provide an easy and fast tool for assessing sulla digestibility.

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