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# Drivers of *Setaria pumila* (Poir.) Roem. et Schult growth and impact on forage quality in lowland Switzerland meadows

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#### 1 Abstract

Invasive grasses (especially Setaria pumila (Poir.) Roem. et Schult) increasingly threaten meadows 2 3 and pastures as a consequence of human impact and climate change. We conducted a study in 2012 and in 2013 to better understand the growing cycle and the influence of S. pumila on forage quality 4 5 of lowland meadows. We observed a rapid increase of S. pumila presence and phytomass on the southern side of the Alps across the growing season. We measured: (i) above-ground phytomass, with 6 a modified Corral-Fenlon method; (ii) botanical composition using the linear point quadrat method; 7 and (iii) grassland chemical composition by NIRs (Near-infrared spectroscopy) analyses. To test the 8 hypothesis that S. pumila summer growth is related to specific climatic conditions, meteorological 9 data (temperature, precipitation and evapotranspiration) were collected from meteorological stations 10 near the study sites. Total phytomass was sorted into S. pumila and other species. We used a 11 Generalized Linear Mixed Models (GLMM) and found the abundance of S. pumila to be inversely 12 correlated with rainfall and the presence of other species, but positively correlated with temperature 13 increase. The increase of S. pumila above-ground phytomass production was linked to a reduction of 14 forage quality. 15

## 16 Keywords

17 Summer grass weeds, Drought, Generalized Linear Mixed Model, NIRs analyses, Forage quality

# 18 Introduction

Ecologists and experts in the biological invasion of meadows and pastures are increasingly interested in better understanding patterns of plant encroachment and mechanisms driving invasive species along the elevation gradient (Alexander et al., 2011). As a genus, *Setaria* represents some of the most problematic weeds that interfere with world agriculture and in other disturbed and managed habitats (Dekker, 2003). In recent decades, C4 weedy summer grasses have increasingly threatened lowland grasslands in Switzerland. Therefore, Nucera et al. (2013) surveyed permanent meadows and lowland pastures from 2009 to 2011 to characterize the forages of a southern Alpine area (Canton Ticino, CH). Results show that the growth of *Setaria pumila* (Poir.) Roem. et Schult corresponds to an increase in
above-ground phytomass production, especially during the summer.

Setaria pumila is native to Europe, but is known throughout the world as a common weed. It 28 is widespread in meadows and in many other anthropic places. Setaria pumila establishment takes 29 place predominantly in disturbed areas such as trampled sites, wheel ruts, heavily grazed areas within 30 paddocks and in dung (Dekker, 2003; James, 2007). Setaria pumila awns can be disseminated within 31 and between farms by machinery, birds and livestock. Setaria pumila seeds are also spread by water 32 and as contaminants in crops and hay. Seeds can remain dormant in the soil for several years (Tozer 33 et al., 2008), can survive in dung in effluents stored in holding tanks and for several years in turf 34 (Masin et al., 2006). These dormant seeds thus potentially provide a source of re-infestation of grass 35 36 swards. Once established in a community, the phenotypic plasticity of individual weedy *Setaria* spp. allows them to adapt to local conditions, maximizing growth, and providing competitive advantages 37 over neighbors (Dekker, 2003). 38

Semi-natural forage resources are very important for pastoral activities in plains and valley 39 floors. To adequately conserve these forage resources, we must better understand the more suitable 40 conditions for the growth of invasive weedy summer grasses, which requires consideration of many 41 environmental, ecological and management factors. The relationship between climate and biological 42 43 invasions has been widely investigated to predict potential invasive plant distribution (Broennimann et al., 2007; Richardson and Thuller, 2007). Weedy summer grasses are generally favored by 44 increasing temperatures and high light intensity. The more efficient fixation of carbon by C4 plants 45 provides competitive advantages for available space and other resources over the less efficient C3 46 species, which are more susceptible to moisture and to heat stress (Tozer et al., 2009). Pastures are 47 vulnerable to the invasion of summer weeds such as S. pumila, largely due to climatic factors that 48 limit the persistence of other species (Tozer et al., 2012). 49

50 Our research was conducted with two main objectives: (i) to determine the principal 51 environmental factors that favour the growth of *S. pumila*, and (ii) to quantify the impact of *S. pumila*  on forage quality. Many studies have emphasized the importance of better understanding *S. pumila* growth effects on meadows and pastures (Fava et al., 2000), especially in New Zealand (Dekker, 2003; James, 2007; Tozer et al. 2008; Tozer et al., 2009; Tozer et al., 2012). To our knowledge, this is the first time that growth data for *S. pumila* above-ground phytomass in southern Alpine areas have been analysed and compared with forage energy content across the growing season.

## 57 Materials and methods

#### 58 *Study species*

Setaria pumila, which has a C4 carbon-fixing pathway, belongs to the Poaceae family. This summer annual grass grows from 20 centimeters to over a meter in height and has mostly hairless stems ranging from green to purple-tinged in color (Lauber et al., 2012). The inflorescence, present from July to the beginning of October, is a stiff, cylindrical bundle of spikelets, from 2 to 15 centimeters long, with short, blunt bristles. *Setaria pumila* prefers bright, low altitude sites (Lauber et al., 2012) with dry soils that are rich in assimilable nitrogen with a slightly acid to neutral reaction (pH 4.5-7.5) (Landolt, 2010).

## 66 Study area

We conducted the study from May 2012 to October 2013 in Canton Ticino (Switzerland), at three different sites (Fig. 1, Table 1): two located in Cadenazzo in the Ticino River plain (Site 1 and Site 2), and one in the municipality of Biasca in the Blenio Valley floor (Site 3). The three study sites were chosen analyzing data collected by AGRIDEA from 2009 to 2011 during the PraMig (Prati Migliori) project on a sample of 20 farms (Nucera et al., 2013). All sites were un-irrigated mowed meadows, they were characterized by a homogenous botanical composition (Appendix 1).

Two sites (site 1, chosen as representative of valley bottom meadows in Canton Ticino, and site 3, representative of upper valley bottom meadows) were representative of *Dactylis glomerata* L. permanent grasslands, one of the four main grassland types in Switzerland, established under fairly

intensive management conditions (Jeangros and Thomet, 2004). Site 2 was a long-term temporary 76 77 meadow converted from an orchard and sown with a grass seed mix (OH-440), notably widespread in the region, with Phleum pratense L. and Poa pratensis L. as dominant species, five years ago. All 78 sites are located in a torrid area (Schreiber et al., 1977) characterized by dry, mild winters and hot 79 summers. In Cadenazzo, where an Agroscope Cadenazzo ACW meteorological station is located, the 80 average temperature and the total annual precipitation resulting from a thirty-year climatic series were 81 11.7 °C and 1802 mm, respectively (Fig. 2a). In Biasca the mean temperature and total annual 82 precipitation were on average 11.9 °C and 1429 mm (mean values from a ten-year climatic series 83 from the Federviti sez. Biasca meteorological station; Fig. 2b). 84

# 85 Sampling design and data collection

96

Surveys were conducted in two plots of 6.5 m<sup>2</sup> (8.7 m x 0.75 m) at each site. Each plot was located 86 in a homogeneous and representative area of the site after a thorough visual estimation of the 87 vegetation. We sampled the botanical composition in all plots every 14 days using a vertical point 88 quadrat method (Daget and Poissonet 1971). We sampled from mid-April at the two Cadenazzo sites, 89 and from mid-May at Biasca, until mid-October for the three sites. For each botanical survey, we 90 identified and recorded 25 plant species intercepts touching a steel needle, along a 7.5 m transect. For 91 each survey, we determined the frequency of occurrence of every species  $(f_i)$  recorded in the transects, 92 93 which is an estimate of species canopy cover (Gallet and Roze, 2001). We calculated Species Relative Abundance (SRA<sub>i</sub>), which was used to detect the proportion of biomass for the different species 94 (Lonati et al., 2009), according to the equation of Daget and Poissonet (1971). 95

$$SRA_i = \frac{f_i}{\sum_{i=1}^{i=n} f_i} \times 100 \ (\%)$$

We assessed herbaceous phytomass and forage production in each plot applying modified Corral-Fenlon method (Corral and Fenlon, 1978; Acutis and Reyneri, 1992; Mosimann, 2011), using two plots instead of four, and 14 days instead of one week between two cuts. After each cut with a sickle bar mower at 5 cm of height, we weighed the above ground phytomass of each plot and dried samples in an oven at 105 °C for 12 hours to determine the sward dry matter. During the two years of study we performed 81 surveys (41 in 2012 and 40 in 2013) to determine botanical composition and above ground phytomass production. We estimated the phytomass of *S. pumila* by multiplying the total above-ground phytomass by its SRA, i.e. the proportion of *S. pumila* in the sward. To assess the proportion between *S. pumila* and other species, we estimated the above-ground phytomass of other species to be the difference between the total phytomass and the estimated value of *S. pumila*.

107 Furthermore, after each cut we randomly collected grass samples (41 in 2012 and 40 in 2013) from the above ground phytomass to assess the variation of the forage quality across the growing 108 season. Samples were cut at 7 cm and were dried in an oven at 60 °C for 12 hours. We ground them 109 110 with a Retsch SR300 mill to pass a 0.25 mm screen, and we performed NIRs analyses with a Büchi NIRFlex N500. The sample were selected from the overall above ground phytomass to represent the 111 proportion of plant species in each site for each cut. For a synthetic expression of herbage quality we 112 113 used Net Energy for Lactation (NEL) as most of the forage of the areas is used as dairy cow feed. The NEL, expressed in MJ, was computed according to Scehovic (1979, 2011). NIRs calibrations 114 were developed by Agroscope and regularly improved by comparison with chemical analyses. Net 115 energy for lactation (NEL) results from the computation of following parameters: organic matter 116 (OM), dry matter (DM), digestible crude protein (DCP), lignin (L), neutral detergent fiber (NDF) and 117 118 digestible organic matter content (DOMD).

To test and quantify the hypothesis that *S. pumila* summer growth is related to specific climatic conditions, meteorological data (temperature, precipitation and evapotranspiration) were collected from two Campbell-CR1000 stations. The evapotranspiration was obtained according to Turc equation (Turc, 1961). For Site 1 and Site 2, we used the meteorological Agroscope station in Cadenazzo and for Site 3 we used the Federviti sez. Biasca station. We calculated thermal time (Tt, °C·day<sup>-1</sup>), total precipitations (TP, mm·day<sup>-1</sup>) and evapotranspiration (ET, mm·day<sup>-1</sup>) for each time interval between two cuts, from the beginning of the growing season in both years. 126 Stastistical analyses

We performed two analyses to determine the principal factors affecting *S. pumila* germination and growth. First, we related the trend of *S. pumila* above-ground phytomass production to climatic conditions, to identify the most favorable conditions, then we related the trend to the presence of other species, to assess the effect of *S. pumila* competition.

We analyzed these relationships using a Generalized Linear Mixed Model (GLMM) with *S. pumila* above-ground phytomass as a dependent variable and with Tt, TP and the phytomass of the other species as independent variables. We excluded variables correlated to a threshold of Pearson coefficient (R) > 0.7. Thus ET was excluded from the analysis because of its correlation to thermal time. In the model a gamma distribution was specified for the continuous dependent variable *S. pumila* phytomass, as normality was not met with Kolmogorov-Smirnoff test (Zuur et al., 2009).

To quantify the impact of *S. pumila* on forage quality, we performed a second GLMM using the variation of forage net energy for lactation as a dependent variable and *S. pumila* above-ground phytomass during the growing season as an independent variable. GLMM was preferred to GLM to check the independence of phytomass measurements against site location (Site 1, Site 2 and Site 3) and year (2012 and 2013), which were considered as random factors. We performed these analyses with R software, version 2.15.3 (R Foundation for Statistical Computing, 2013) using "glmmADMB" package (SVN revision 231).

### 144 **Results**

The proportion of *S. pumila* changed at the study sites during the growing season in both years. *Setaria pumila* maximum abundance occurred at the beginning of summer (SRA was 46%, 44%, and 24% in Site1, 2, and 3 respectively in 2012; SRA was: 41%, 46%, and 10% in Site 1, 2, and 3 respectively in 2013).

Climatic data (Fig. 3a and 4a) measured across the growing season at all sites showed that 2013
precipitation from the beginning of April to the end of May was greater than 2012 precipitation during

the same period. Many drought periods occurred from June to August of both years, during the *S*.
 *pumila* growing season.

Growing season forage production data show that Site 1, the most productive, produced an 153 average of 15300 kg of DM·ha<sup>-1</sup>·y<sup>-1</sup>, while Site 2 produced 13400 kg of DM·ha<sup>-1</sup>·y<sup>-1</sup>, and Site 3 had 154 the lowest production level with 8200 kg of DM·ha<sup>-1</sup>·y<sup>-1</sup>. We observed an anomalous summer 155 production peak of the above-ground phytomass (Figs. 3b, 4b) that in 2012 measured 97 kg DM·ha<sup>-</sup> 156 <sup>1</sup>·d<sup>-1</sup> at Site 1 (Fig. 3b), 103 kg DM·ha<sup>-1</sup>·d<sup>-1</sup> at Site 2 (Fig. 3c), and 71 kg DM·ha<sup>-1</sup>·d<sup>-1</sup> at Site 3 (Fig. 157 4b). In 2013 the same phenomenon occurred, with 96 kg DM·ha<sup>-1</sup>·d<sup>-1</sup> at Site 1 (Fig. 3b), and 110 kg 158 DM·ha<sup>-1</sup>·d<sup>-1</sup> at Site 2 (Fig. 3c). In 2013 at Site 3 the production peak, 36 kg DM·ha<sup>-1</sup>·d<sup>-1</sup> (Fig. 4b), was 159 160 reduced compared with the previous years' peak (Fig. 4b). At all sites in both years, the total aboveground phytomass production peak corresponded temporally and by weight to the S. pumila above-161 ground phytomass production peak. 162

We observed a reduction in forage quality, expressed in MJ of NEL, in both years and at all sites corresponding to the highest *S. pumila* above-ground phytomass production (Figs. 3b, 3c; 4b).

At all sites we observed NEL values of around 6.5-7.0 MJ (Figs. 3b, 3c, 4b) at the beginning 165 of the growing season, which decreased considerably to below 5.0 MJ with the ingress and the 166 development of S. pumila in the summer. At Site 3 in 2013 we observed a stable trend in the net 167 energy for lactation throughout the growing season probably due to an absence of S. pumila (Fig. 4b). 168 The results of the first GLMM (Table 2) show that S. pumila above-ground phytomass was 169 inversely correlated with other species above-ground phytomass and to rainfall, but was positively 170 correlated to temperature increase. The result of the second GLMM (Table 3), which was statistically 171 significant, highlights that forage quality was inversely related to S. pumila phytomass production. 172

## 173 Discussion

At all sites in both years, the *S. pumila* above-ground phytomass production peak corresponded to the total above-ground summer phytomass production peak when, according to meteorological data,

drought and high temperatures occurred. The observed differences in herbage yield and in S. pumila 176 177 abundance (especially at Site 3) were probably due to differences in climatic conditions, in particular due to spring precipitation, which was greater in 2013 than in 2012. However, small climatic changes 178 had little impact on S. pumila colonization capacity (Dekker, 2003). The successful establishment of 179 180 all Setaria spp. can be ascribed to their intimate evolutionary relationship with humans, disturbance, agriculture, and land management and to their ability to adapt rapidly to local conditions (Dekker, 181 182 2003). The relationship between S. pumila phytomass and climatic parameters confirms that results reported in other countries also apply to the Southern Alps. In fact it was demonstrated that S. pumila 183 cover increases after severe droughts, while the proportion of less competitive, good forage species 184 185 decreases (Tozer et al., 2012). Bare ground is essential for germination and establishment of 186 colonizing species (Bullock, 2000; Tozer et al., 2012) such as weedy annual grasses, among which S. pumila is the dominant species (Tozer et al., 2009). The preservation of good forage species, which 187 results in a competitive sward, is essential to minimize the amount of bare ground available for 188 colonization by weedy annual grasses (Tozer et al., 2009). Pastures effective in preventing invasion 189 190 of weedy annual grasses should have few gaps available for colonization by invasive plants (Tozer et al., 2012). 191

192 The reduction in pasture quality during the two years of study was significantly dependent on 193 S. pumila, but was likely not related to the phenological stage of the species, as plants were kept at an early phenological stage by Corral-Phenlon mowing every two weeks. The forage net energy for 194 lactation of meadows and pastures was strongly reduced by the presence of S. pumila, which produces 195 196 forage characterized by indigestible organic matter (Tozer et al., 2012). Ingestion of S. pumila also causes mouth ulceration and reduced forage intake, moreover decreased milk production have been 197 198 observed when dairy cattle are fed with hay containing a high content of *S. pumila* (Fava et al., 2000). Invasion by S. pumila and loss of good forage species negatively affects farm economy (Tozer et al., 199 2008). Weed management and other human activities are fundamental selection forces affecting 200 herbaceous invasive species. The traits and adaptations that surviving weedy Setaria spp. plants 201

acquire in this struggle will continue to be urgent problems confronted by land managers (Dekker,
203 2003).

#### 204 Conclusions

These first analyses of S. pumila growth data outline the processes that affect its abundance and the 205 impact on forage quality of temporary and permanent meadows. Our results demonstrate that the 206 large increase of sward above-ground phytomass during the summer can be attributed to S. pumila, 207 when this species is probably more competitive than other species under drought conditions and high 208 temperatures, due to its C4 carbon fixation pathway. This research raises concerns about the spread 209 of weedy summer grasses to higher elevations as a consequence of climate warming and human 210 pressure in mountain systems. Monitoring the existing population is important, but preventing the 211 introduction of invasive species is a more urgent goal, paying special attention to herbaceous species 212 (such as Setaria spp.), which can flower over a short period and later in the season. 213

214 The extent to which the species realizes its potential distribution in dairy pastures will depend on the extent to which its seeds are dispersed. Setaria pumila is dispersed mainly by human activities 215 and on-farm biosecurity measures will be required to limit its spread. Trying to limit S. pumila growth 216 with chemical methods in meadows and pastures is unfeasible, but improving soil fertility, 217 maintaining a high pH (close to 6) and modifying grazing and mowing management could enhance 218 219 the competition of more desirable species. However, to turn the research results into practice, farmer training is essential. Pasture species mixtures and management practices that impede the ingress of S. 220 *pumila* have to be investigated in order to avoid the loss of good forage species and the reduction of 221 222 the farm yield.

Our study looked at herbaceous meadow vegetation, which is normally mowed, but we recommend further analyses of *S. pumila* and other C4 weedy summer grasses on the southern side of Alps, that consider all of the effects of management practices on meadow botanical composition.

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- 298

# **Table 1** – Sites characteristics

	Coordinates <sup>1</sup>	Municipality <sup>2</sup>	Elevation <sup>3</sup>	Dominant species <sup>4</sup>
Site 1	46°9'37'' N	Cadenazzo	202 m	Dactylis glomerata /
	08°56'0'' E			Lolium multiflorum
Site 2	46°9'37'' N	Cadenazzo	203 m	Phleum pratense / Poa
	08°56'3'' E			pratensis
Site 3	46°24'25'' N	Biasca	370 m	Dactylis glomerata / Poa
	08°58'27'' E			annua
$^{1}$ Coord	inates of the site	in UTM WGS 8	$4 \cdot {}^2$ Locality of	the site <sup>3</sup> Elevation of the site (m

<sup>1</sup> Coordinates of the site in UTM WGS 84; <sup>2</sup> Locality of the site; <sup>3</sup> Elevation of the site (meters above sea level); <sup>4</sup> Dominant species before *S. pumila* appearance.

- **Table 2** GLMM summary with *S. pumila* phytomass as dependent variable related to other
- 306 species phytomass, Tt and TP

GLMM parameters	Estimate	Std. error	Signif. <sup>1</sup>
Intercept	4.573	0.641	***
Other species phytomass (kg DM)	- 0.986	0.305	**
Tt (°C*day)	2.170	0.748	**
TP (mm*day)	- 1.435	0.321	***
<sup>1</sup> ·***'P<0.001, ·**'P<0.01			
Table 3 – GLMM summary with NFI	value as den	endent variah	e related :
	GLMM parameters         Intercept         Other species phytomass (kg DM)         Tt (°C*day)         TP (mm*day)         1 '***'P<0.001, '**'P<0.01	GLMM parametersEstimateIntercept $4.573$ Other species phytomass (kg DM) $-0.986$ Tt (°C*day) $2.170$ TP (mm*day) $-1.435$ $^{1}$ ****'P<0.001, '**'P<0.01	GLMM parameters         Estimate         Std. error           Intercept         4.573         0.641           Other species phytomass (kg DM)         - 0.986         0.305           Tt (°C*day)         2.170         0.748           TP (mm*day)         - 1.435         0.321

312 ground phytomass

GLMM parameters	Estimate	Std. error	Signif.1
Intercept	1.83 <sup>e</sup>	1.33 <sup>e-2</sup>	***
S. pumila phytomass	- 1.57 <sup>e-4</sup>	2. <sup>06 e-5</sup>	***
<sup>1</sup> ·***'P<0.001			



318 Fig. 2 – Monthly average temperature and total precipitations of climatic series in [a] Cadenazzo

319 and in [b] Biasca



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Fig. 3 – Comparison of 2012 and 2013 growing season data corresponding with the mowing of each plot, expressed in days of the year between: (a) Cadenazzo meteorological data, histogram for weekly average precipitation and hatched line for daily average temperature; (b) Site 1 data; (c) Site 2 data. The double line curve represents the daily above-ground phytomass production, the continuous grey line symbolises S. pumila above-ground phytomass and the dotted black line indicates nutritive value, expressed in MJ of net energy for lactation (NEL).



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Fig. 4 – Comparison of 2012 and 2013 growing season data corresponding with the mowing of each plot, expressed in days of the year between: (a) Biasca meteorological data, histogram for weekly average precipitation and hatched line for daily average temperature; (b) Site 3 data. The double line curve represents the daily above-ground phytomass production, the continuous grey line symbolises S. pumila above-ground phytomass and dotted black line indicates nutritive value, expressed in MJ of net energy for lactation (NEL).



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