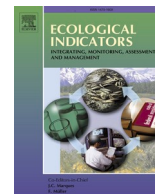


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Monitoring biomass in two heterogeneous mountain pasture communities by image based 3D point cloud derived predictors

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ABSTRACT

Primary productivity is a robust indicator of ecosystem functioning because of its close relationships with the stability of the ecological systems. In ecological research, the above ground biomass (AGB) is the most commonly used proxy of primary productivity. However, despite their ecological relevance, the estimates of primary productivity are not addressed by current protocols for monitoring the conservation status of the habitats of Community interest. In this paper, we analyse the accuracy of AGB measurements obtained by image-derived 3D reconstructions of two contrasting mountain grasslands listed as habitats of Community interest in the Annex I of the Habitats Directive. More specifically, we compared the accuracy of the AGB estimates provided by four models, based on four different predictors (height, volume, volume adjusted, and cover volume), in order to evaluate their robustness against within- and between-community heterogeneity. Our study revealed that AGB measures computed from 3D vegetation reconstructions can be an effective way to evaluate primary productivity in herbaceous communities with complex structure and composition patterns. In particular, the vegetation height showed to have the highest correlation with direct AGB measurements. However, the vegetation volume, once adjusted by the coefficient of density, resulted to be the most effective proxy due to the lowest error level. Therefore, such a parameter could be routinely used as a non-destructive indicator for monitoring habitats of particular conservation concern. As a major limitation for this approach, we detected some loss of predictivity power at very low productivity rates.

1. Introduction

Pasture and grassland communities occurring in Mediterranean mountain areas possess an extraordinary floristic richness, and qualify as relevant biodiversity hotspots (Wilson et al., 2012). To date, these communities are included among the most threatened ecosystems due to the environmental variations that the ongoing global change is promoting in southern European mountain areas (Schröter et al., 2005). Therefore, understanding how such species-rich communities cope with environmental changes is needed for building up reliable expectations and effective conservation strategies. In this framework, using reliable indicators for monitoring community responses to environmental variations represents a crucial requirement. Primary productivity is considered the indicator that best reflects changes in ecosystem functioning because of its connection to ecosystem stability and resistance (de Bello et al., 2010). Indeed, plant productivity responds rapidly to changes in resource availability caused by variations in water, nutrients,

and disturbance regimes (Herrick et al., 2005). Moreover, primary productivity closely depends on biodiversity, as biodiversity variations often result in a reduction of productivity (Li et al., 2015; Tilman, 1996). Therefore, monitoring primary productivity is essential to understand how environmental variations affect ecosystems. Especially, under the global biodiversity crisis, monitoring primary productivity can help us to understand how the ongoing biodiversity variations can impact the functioning of species-rich ecosystems like the oro-Mediterranean grasslands.

Above ground biomass (AGB) is the most visible of the five carbon pools in terrestrial ecosystems (Ravindranath and Ostwald, 2008), and is widely used in ecological studies as a proxy of primary productivity. In grasslands, AGB estimates can help in understanding the impact of biophysical and ecological processes on ecosystem productivity (Loreau and Hector, 2001; Tilman et al., 2006) and quantifying the effect of an array of biotic and abiotic factors on temporal and spatial productivity changes (Augustine, 2003; Frank and McNaughton, 1991; Knapp and

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Smith, 2001). For such reasons, AGB is a reliable parameter for a wide range of ecological applications concerning grasslands, including habitat monitoring (McNaughton, 1985), grazing (Trotter et al., 2010) and fire management (Trollope et al., 1996), and carbon storage (Tilman et al., 2006). The long-term AGB monitoring in grassland communities can provide an effective tool to improve land management strategies, especially with regards to maintenance and/or restoration of composition, structure and diversity of pasture plant communities (Folts-Zettner et al., 2011).

Over time, ecologists have applied several techniques for studying AGB, but the most accurate method requires cutting, drying and weighing of biomass samples (Catchpole and Wheeler, 1992; López-Díaz and González-Rodríguez, 2011). In spite of its large use, this approach has some relevant limitations. Indeed, it is time and labour demanding (Brummer et al., 1994; Catchpole and Wheeler, 1992; Wachendorf et al., 2018) and allows for studying only limited surfaces. Moreover, because the plants are physically removed, the estimate cannot be repeated over time (Cooper et al., 2017; Frank and McNaughton, 1991; Passalacqua et al., 2019). Due to such limitations, the application of this destructive method appears unsuitable in protected areas and in programs of repeated surveying as required by the monitoring protocols for community conservation (Passalacqua et al., 2019). On the contrary, visual estimate is the most feasible approach to evaluate AGB (Ónodi et al., 2017; Redjadj et al., 2012), even when associated to canopy height (Axmanová et al., 2012), cover (Röttgermann et al., 2000), or calibrated by double sampling (Ebrahimi et al., 2008) and reference unit method (Boyda et al., 2015). However, the individual ability to visual AGB estimates is greatly variable. Consequently, both precision and accuracy of such evaluations can be low, variable, and difficult to define, limiting the value of the obtained AGB data (Herrik et al., 2009; Tucker, 1980).

To overcome the limitations of these measurements, ecologists tried to develop alternative methods for non-destructive AGB estimations (Catchpole & Wheeler, 1992; Kumar et al., 2015; López-Díaz and González-Rodríguez, 2011 and therein literature). For instance, disc pasture meter (Santillan et al., 1979), point intercept method (PIM, Jonasson, 1988), and visual obstruction methods (Robel et al., 1970) have often been used in vegetation studies as reliable linear proxies of plant biomass. Such methods can provide moderate to good biomass predictions. Working in managed pastures, which are characterised by a homogeneous structure and species composition, accurate biomass estimates can be achieved by using manual instruments (Byrne et al., 2011; Duru and Bosquet, 1992; Earle and McGowan, 1979; Gonzalez et al., 1990; Griggs and Stringer, 1988; Michell and Large, 1983; Robel et al., 1970). On the contrary, when applied in highly diversified grasslands, these methods result less effective, producing sometimes quite contrasting outcomes. For instance, the cover estimation approach produced subjective and little repeatable results also in managed grasslands, (Godínez-Alvarez et al., 2009; Greig-Smith, 1983; Tucker, 1980; Wilson, 2011), performing less effective predictions than point and line intercept methods. Instead, in wild herbaceous communities, there was no evidence that visual cover estimation is less accurate than other approaches for biomass estimation, including point intercept method, field spectroscopy, plate meter, and 3D quadrat method (Ónodi et al., 2017). Furthermore, such methods are associated with a moderate to high experimental error, because relationships between AGB and AGB predictors depend on numerous factors that can interact mutually (López-Díaz and González-Rodríguez, 2011). Besides, prediction models based on field measurements are expensive in terms of time and field efforts (Byrne et al., 2011). To improve sampling capacity and precision further more complex electronic instruments have also been developed, such as the electronic capacitance meter (Fletcher and Robinson, 1956) and the sonic sward stick (Hutchings et al., 1990). However, as reported by Murphy et al. (1995), electronic capacitance meter readings are affected by water in vegetation; despite using standardised equations, they are not representative of different conditions and situations (Frame, 1993).

As far as the remote sensing approach is concerned, this resulted to be an effective way to estimate AGB in grasslands on a regional scale, by allowing large-scale biomass mapping (Li et al., 2013). On the other hand, working at a lower scale, the AGB estimates provided by remote sensing can be less effective if compared to manual instruments (Ónodi et al., 2017; Wigley et al., 2019). Moreover, the performance of remote sensing can be affected by the structure of the study area and by the nature of the remotely sensed data (Kumar et al., 2015; Todd et al., 1998). Finally, it must be considered that the remote sensed data generally require field measurements for calibration and validation (Wallace et al., 2017).

Among the most-promising methods for non-destructive AGB estimation, the techniques based on three-dimensional (3D) community reconstruction are receiving a growing interest, because they permit to obtain biomass estimates without using allometric information (Calders et al., 2015). For instance, Terrestrial Laser Scanning (TLS) techniques were frequently used to obtain 3D point clouds to produce three-dimensional reconstructions of woody vegetation. Some authors (Hütt et al., 2014; Owers et al., 2018; Schulze-Brüninghoff et al., 2019; Tilly et al., 2014) used TLS based 3D point clouds of herbaceous communities to assess grass height and volume to perform estimations of AGB, by finding out a meaningful correlation between model measurements and destructively harvested data. However, image-based 3D reconstruction (Structure-from-Motion, SfM) (Snaveley et al., 2008) appears an effective low-cost methodology alternative to laser-based systems (Westoby et al., 2012). Recent comparisons between SfM- and TLS-based AGB estimates showed that both methods allow performing an accurate estimation of the surface biomass in pasture communities. However, TLS is much more demanding in terms of expenses and expertise (Cooper et al., 2017; Kumar et al., 2015; Wallace et al., 2017). SfM was applied to create grassland 3D reconstructions (Cooper et al., 2017; Passalacqua et al., 2019; Wallace et al., 2017; Wigley et al., 2019), even by the use of unmanned aerial vehicle (UAV) (Bareth and Schellberg, 2018; Grüner et al., 2019; Lussem et al., 2019), reporting good correlation values to AGB. In addition, Passalacqua et al. (2019) demonstrated that SfMs allows to obtain reliable diachronic estimates of fine-scale vegetation productivity (i.e. volume-based AGB variations) in highly diverse communities like oro-Mediterranean pastures.

The effectiveness of image-based techniques for diachronic AGB measurements suggests that this non-destructive approach can be a valuable tool for fine-scale monitoring of spatial-temporal variations of community primary productivity. Therefore, such an approach could be successfully integrated in conservation monitoring programs. For instance, the Habitats Directive 92/43/EEC and the Nature 2000 network represent the most relevant framework for biodiversity conservation in the European Union. The Habitats Directive demands rigorous conservation policies to preserve wild habitats and species in a favourable conservation status (European Council Directive 92/43/EEC of 21 May 1992). As a core point in the promoted strategy, art. 17 of the Habitats Directive requires a periodical assessment of the conservation status of the habitat types listed in its Annex I, which includes a wide array of herbaceous communities. The assessment consists of collection of data regarding specific habitat parameters according to European (European Commission, 2018) and related national guidelines (Angelini et al., 2016). To date, monitoring guidelines and protocols for assessing the conservation status of habitats of Community interest do not include estimations of primary productivity. Nevertheless, the availability of a non-destructive, low-cost, and effective methodology would allow integrating this crucial ecological indicator to monitor functioning and dynamics of pastures and grasslands of Community interest.

Therefore, in this paper we analyse the accuracy of AGB measurements obtained by image-derived 3D reconstructions of two contrasting mountain communities listed as habitats of Community interest in Annex I of the Habitats Directive. Substantially, our framework aimed to identify the best protocol for using SfMs to monitor the primary productivity of herbaceous communities of conservation interest. To this

end, we identified three main goals: 1) performing AGB predictive models based on four different predictors, 2) comparing the accuracy of the AGB estimations provided by the different models, and 3) evaluating the robustness of predictors against within- and between-community heterogeneity.

2. Materials and methods

2.1. Study area

The study was carried out in three working areas in the Pollino National Park (S-Italy), the largest protected area in Italy (Fig. 1). The Pollino National Park includes the highest mountain tops of the S-Apennines, and it is positioned in the middle of the Mediterranean Basin. Overall, the local climate shows the typical seasonal contrast between hot dry summer and mild wet winters. To test the experimental protocol under different productivity regimes and represent differences in elevation and ecological context (i.e. xerophile vs. mesophile), three study areas were selected: Serra delle Ciavole (hereafter SC: N 39.92508°, E 16. 20968°; elevation: 1900 m a.s.l), Piano di Ruggio (hereafter PR: N 39.91197°, E 16.13053°; elevation: 1600 m a.s.l), Monte Serra (hereafter MS: N 39.84804°, E 16.09311°; elevation: 1400 m a.s.l.). The elevation is a major driver of regional climatic variations; therefore, it can strongly affect patterns of plant productivity. In the study system, climatic conditions ranged from meso-Mediterranean (at

MS), where summer drought is slightly reduced, to mid-continental one (at SC), where the winter is very cold and a long-lasting snow-cover lies from November to May. Grazing is a further driver of plant productivity showing a marked spatial variability in the Pollino National Park (Gargano et al., 2012). Although all study areas are subject to grazing, the source and intensity of grazing are variable among sites. SC has the highest grazing pressure, mainly due to horses and cows; PR undergoes a middle to high pressure caused by bovine grazing; finally, MS is subjected to a moderate bovine and ovine grazing. As far as the ecological context is concerned, each work area includes both xerophile pastures and mesophile grasslands, which differ for striking variations of floristic structure and composition.

Xerophytic communities are characterized by a discontinuous vegetation cover, due to the rocky nature of the substrate, showing poorly developed soils and high drainage (Fig. 2a). The dominant species include *Armeria canescens* Boiss., *Bromopsis erecta* (Huds) Fourr., *Festuca circummediterranea* Patzke, and *Poa alpina* L. These communities qualify as habitat of Community interest, being listed in the Annex I of the Habitats Directive with the code '6210 *Semi-natural dry grasslands and scrubland facies on calcareous substrates (Festuco-Brometalia) (*important orchid sites)*'. This habitat hosts a high plant diversity, including many orchid species, and its conservation depends on the maintenance of traditional pastoral activities. In the S-Apennines, the mesophytic grasslands are rarer than the previous ones, because they are confined to high-mountain areas showing a flat surface covered by deep

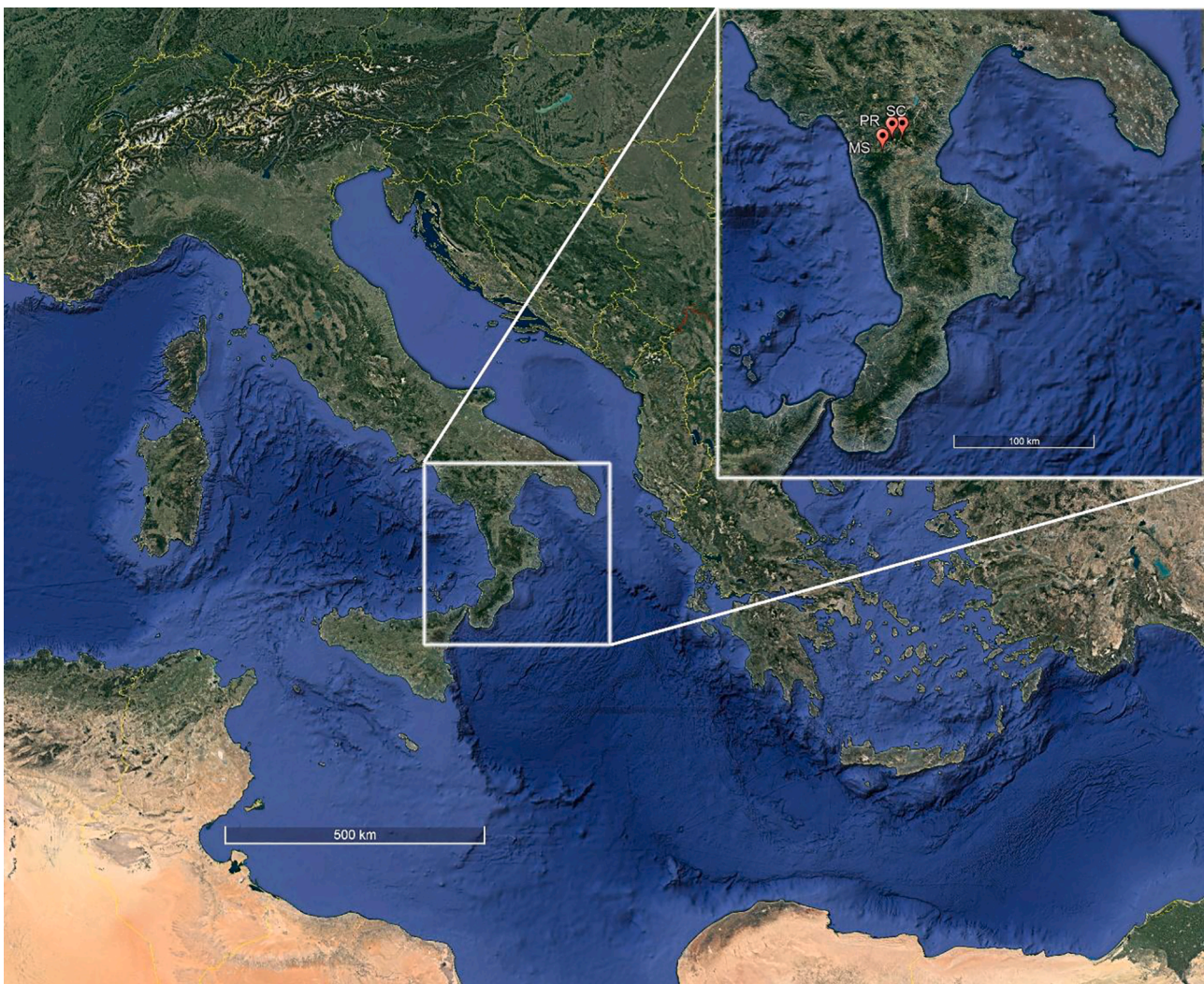


Fig. 1. Map showing where are the study areas in the Mediterranean area and in the Calabria region: Monte Serra (MS), Piano Ruggio (PR), and Serra Ciavole (SC).



Fig. 2. Images of the xerophytic (a) and the mesophytic (b) pastures.

and loamy soils (Bonin, 1972; Gargano et al., 2010) (Fig. 2b). The dominant species include *Achillea millefolium* L., *Agrostis capillaris* L., *Cynosurus cristatus* L., *Dactylis glomerata* L. subsp. *hispanica* (Roth.) Nyman, *Festuca microphylla* (St.-Yves ex Coste) Patzke. In addition, such communities host some rare endemic taxa (e.g., *Plantago media* L. subsp. *brutia* (Ten.) Arcang.), and species close to their range border (e.g. *Gentiana lutea* L.) (Gargano et al., 2017). These formations belong to the Meo-Asphodeletum association and fall within the habitat of Community interest coded in the Annex I of the Habitats Directive as ‘6170: *Alpine and subalpine calcareous grasslands*’.

To carrying out field work, a sampling site was defined in each community type in all the three areas. Each sampling site consisted in 4 homogeneous plots of 1 × 1 m; plots were defined and delimited by a mobile tubular structure. All the field work was carried out in the period from June to July 2018 and consisted of a total of 24 plots.

2.2. AGB estimation protocol: major steps

The protocol to obtain and test AGB estimates encompassed the following 5 steps: 1. Image acquisition and 3D reconstruction; 2. AGB predictors; 3. AGB sampling; 4. Patterns of AGB and AGB predictors; 5. Analysis of model performance.

2.3. Image acquisition and 3D reconstruction

Image acquisition and processing followed the method used in Passalacqua et al. (2019). To take images of the plots, a GoPro Hero 4 Silver model was installed on the top of a 1 m pole equipped with a Feiyu FY-G5 gimbal device to reduce pitch and roll movements and stabilize the acquisition. The camera was set in video mode, at a resolution of 1080p at 60 frames per second (fps) and field of view of 180°. The camera network planned to survey the plots consisted in open loop strips taken at about 10–15 cm above the maximum vegetation height. The camera was in downward-looking position and was moved horizontally right–left and left–right on overlapping strips along straight lines, ensuring a sidelap >30%. The occluded areas, not visible in downward-looking filming video, were acquired using oblique poses. Image acquisition requires a trained operator, because the camera has to be moved slowly to avoid blurred frames, which would reduce the 3D reconstruction quality. Standardization of image acquisition is an open question we are working to. To improve image quality, during the acquisition the plot was covered by a tent (Quechua, 2 s 3 XL fresh & black) for minimizing disturbance due to wind and intense light contrast. About 5 videos of 1 min for each plot were obtained, and all videos were fragmented into still frames by using Adobe After Effects 2018 software. After this

process, an extraction ratio of 3/30 fps was obtained, with an average overlapping rate between consecutive images of 60%. Subsequently, the image quality was manually checked by using Adobe Bridge 2019, and 450 frames resulted to be adequate for 3D reconstructions. The Agisoft PhotoScan Pro package was then used for obtaining the models by SfM (Snively et al., 2008). An algorithm similar to Scale Invariant Feature Transform (Lowe, 1999) was applied to orient the images and to build a sparse 3D reconstruction. Besides, a Multi-View Stereo algorithm was used to produce a dense 3D point cloud from the refined intrinsic orientation and ground-referenced camera exterior orientation. 3D-point clouds were processed using the CloudCompare 2.8.1 software. Although the plots were on a nearly flat surface (slope $< 5^\circ$), a planar representation of the ground surface was defined using the Euclidean coordinates of the four vertices of a 1×1 m tubular (0.02 m \varnothing) structure lying on the ground as a reference. These four vertices were used as Ground Control Points (GPCs). A reference geometric entity, with standard dimensions ($1.1 \times 1.1 \times 0.05$ m), was created as laying with the upper surface on the plane at $z = 0$ of the virtual space. The four lower vertices of the box were selected as reference to align the GPCs of the point cloud. In this way, the vertices of the plots assumed a coordinate $z = -5$. Point clouds were cleaned to remove all points outside the perimeter of the considered plot. Finally, it was performed a removal of stones and bare soil which could significantly influence volume calculation. The used softwares do not require special skills, just some basic knowledge of image and 3D vector editing (i.e. GIS), and a few training days are enough to learn and apply procedures for 3D point cloud reconstruction and processing.

2.4. AGB predictors

The two most common metrics used to predict above ground biomass are community height and cover (e.g., Axmanová et al., 2012; Flombaum & Sala, 2007; Gutierrez & Aguilera, 1989; Ónodi et al., 2017; Pottier & Jabot, 2017; Röttgermann et al., 2000). These measures can be combined to calculate the volume as a predictor of above-ground vegetation biomass (Grinath, 2019; Grinath et al., 2015; Gutierrez and Whitford, 1987; Hirata et al., 2007; Penderis and Kirkman, 2014). Accordingly, 3D reconstructions allow for a detailed measure of community height and cover, computing a direct and much more accurate measure of community volume. Both height and volume involve the creation of a raster grid by partitioning the x, y plane of the point cloud into square cells. A normalized vegetation height was then determined based on the height of the point cloud above the cells in the identified ground surface, and the volume as the product of the cell area and the cell height. In this study, height and volume were defined using the following reference parameters: cell grid size of 1 mm, and the minimum height. Indeed, previous work indicated that such parameters offer the best setting for volume estimation (Passalacqua et al., 2019). Height (H) was computed as the average height of the cells, and volume (V) was determined as the product of the cell area and the attributed height.

As showed by Passalacqua et al. (2019), the volume of each plot depends on point cloud density, so that a coefficient of density (C_p) was applied to make the measured volumes comparable. The coefficient of density was defined as:

$$C_p = \rho_{\max} / \rho \quad (1)$$

where ρ_{\max} was the value of the maximum point cloud density of all derived point clouds, while ρ was the density of the point cloud (Passalacqua et al., 2019). The coefficient of density (C_p) was applied to adjust the volume values (V) in relation to variation due to different point cloud densities. In fact, the adjusted volume (V_{adj}) can be defined as

$$V_{\text{adj}} = C_p V \quad (2)$$

where V was the volume of a sampled plot (Passalacqua et al., 2019).

Another way to compute the vegetation volume consists in multiplying average height (H) \times vegetation cover (Grinath, 2019). In 3D reconstructions, the vegetation cover can be computed by multiplying the grid cell surface per the number of cells that include at least a point on the z axis. At 1 mm cell size, cover could be underestimated depending on the 3D cloud density, because the vegetation surfaces result filled with gaps where no point is over a cell. For this reason, we measured the vegetation cover setting cells at 5 mm, reducing the chance that a cell was without any point over. Then, we determined the predictor cover volume (V_{hc}) as average height (H) \times vegetation cover (C):

$$V_{\text{hc}} = HC \quad (3)$$

Cover volume (V_{hc}) represents the traditional way to estimate vegetation volume. Conceptually, it is obtained by a geometrical computation of volume; additionally, as it is based on the average vegetation height, cover volume is not affected by point cloud density.

We used millimetres (mm) as a unit measure for H, and cube decimetre (dm^3) as a unit for volumes.

2.5. AGB sampling

All sampling units were also subjected to traditional estimations of AGB. After image acquisition, the AGB was removed (up to 5 cm from the ground surface) in all the 24 plots. Fresh samples were preserved in nylon bags to be processed in laboratory within 24 h from field sampling. In laboratory, AGB fresh samples were weighted by a digital balance (resolution = 0.01 g) to obtain fresh weight measurements (FW); afterwards, they were dried in oven at 90° for 48 h. After drying, the samples were weighted to measure dry weight (DW). The estimations of FW and DW were then expressed in g/m^2 .

2.6. Patterns of AGB and AGB predictors

Data on biomass (FW and DW), point clouds (density, ρ ; and coefficient of density, C_p) and predictors (H, V, V_{adj} , V_{hc}) were subjected to univariate statistics to determine average value (μ), range (minimum, Min, and maximum, Max), and variation (coefficient of variation, CV). Differences between communities and among sites were evaluated. The t and F -test (with Tukey-Kramer *post-hoc* pairwise comparisons), or Welch test in the case of unequal variance (with Mann-Whitney pairwise *post-hoc* tests, and Bonferroni corrected p value), were used to test differences in average values when variables fitted the normal distribution (Shapiro-Wilks test). The Mann-Whitney test for equal medians was used when normality was not fitted even when variables were transformed. Fligner-Kileen test was applied to test differences in coefficient of variation.

2.7. Analysis of model performance

The best model was identified based on the assessment of the prediction power of the four linear models considering biomass as dependent variable and the parameters H, V, V_{adj} , V_{hc} as predictors. Relationships between predictors and biomass were evaluated using ordinary least square linear regression. Several parameters were evaluated to assess the model fitting: general performance of the model (coefficient of determination, R^2 ; root-mean-square deviation, RMSE; and relative root-mean-square deviation, $\text{rRMSE} = \text{RMSE}/\text{DW}$ mean value), fitness to linear regression assumptions (influencers: maximum Cook distance, DC_{max} ; residuals normal distribution: Shapiro-Wilk test, W ; residuals heteroscedasticity: Breusch-Pagan test, LM), and slope (a) and intercept (b) coefficients statistics (value, standard error, and p -value). Leave-one-out cross validation was used to test model performance (R^2_{cv} , and RMSE_{cv})

Besides, models were compared by the inspection of residuals. First, we analysed the relationships between residuals and biomass by linear regression. A meaningful relationship would lead to a weakness in the

model, indicating that other parameters are required to produce a more reliable estimate of biomass. Then, the weight of residuals (RW) on the measured value of biomass was calculated by the ratio of the residuals on biomass:

$$RW = \text{residual}/DW \quad (4)$$

RW measures whether DW is over- or underestimated; a negative RW value would indicate a DW underestimation, while a positive one would mean a DW overestimation.

Furthermore, the absolute RW value was calculated:

$$RW_{\text{abs}} = |\text{residual}|/DW \quad (5)$$

The RW_{abs} is a measure of the error level, as it shows how large is the residual in respect to the predicted variable. Accordingly, low RW indicates that the estimated biomass value is highly comparable to the measured value, whereas high RW indicates a relatively weak predictive power of volume versus biomass. A non-linear model was applied to assess the relationship between RW_{abs} and biomass and the model function was selected by the lower Akaike information criterion (AIC). RW_{abs} at 0.1, corresponding to an error level of 10% in estimating the biomass, was used to assess the accuracy of the models. Differences in RW and RW_{abs} between communities and among sites were evaluated by the same statistics illustrated in *Patterns of AGB and AGB predictors*.

All the statistical analyses were performed using the software packages Data Desk 6.3.1. (Data Description, Ithaca, New York, USA) and

PAST 4.01 (Hammer et al., 2001).

3. Results

3.1. Patterns of AGB and AGB predictors

Point clouds density ($\mu = 761034$; 1,811,775–222,584 points/m²) provided a good representation of the vegetation structure (Fig. 3). However, the coefficient of density ranged between 1 and 8.14 ($\mu = 3.79$), showing that the quality of point clouds varied significantly among the 3D reconstructions. Accordingly, although the point cloud average density did not vary between communities ($P = 0.66$); among the investigated sites, the 3D-models made at SC revealed a best quality ($\mu = 1409345$; $P < 0.05$).

Biomass harvested in the 24 plots ranged between 1.56 g and 1672.65 g for FW ($\mu = 253.62$), and between 0.52 g and 413.04 g for DW ($\mu = 76.86$) (Table 1); in addition, FW and DW showed a comparable variation (CV = 162, and 133, respectively; $P > 0.5$) (Table 1). FW biomass was highly related to DW biomass ($r^2 = 94.8\%$, $P < 0.0001$); therefore, hereafter we will consider only data on DW.

Average biomass was higher in mesophytic samples than in xerophytic ones (119.2 g and 39.5 g, respectively), but this between-community difference was not significant ($P > 0.05$) (Table 1). Among sites, DW had the lowest average value in SC ($\mu = 4.29$ g), marking a striking difference with PR ($\mu = 162.5$ g, $P < 0.005$), and from MS ($\mu =$

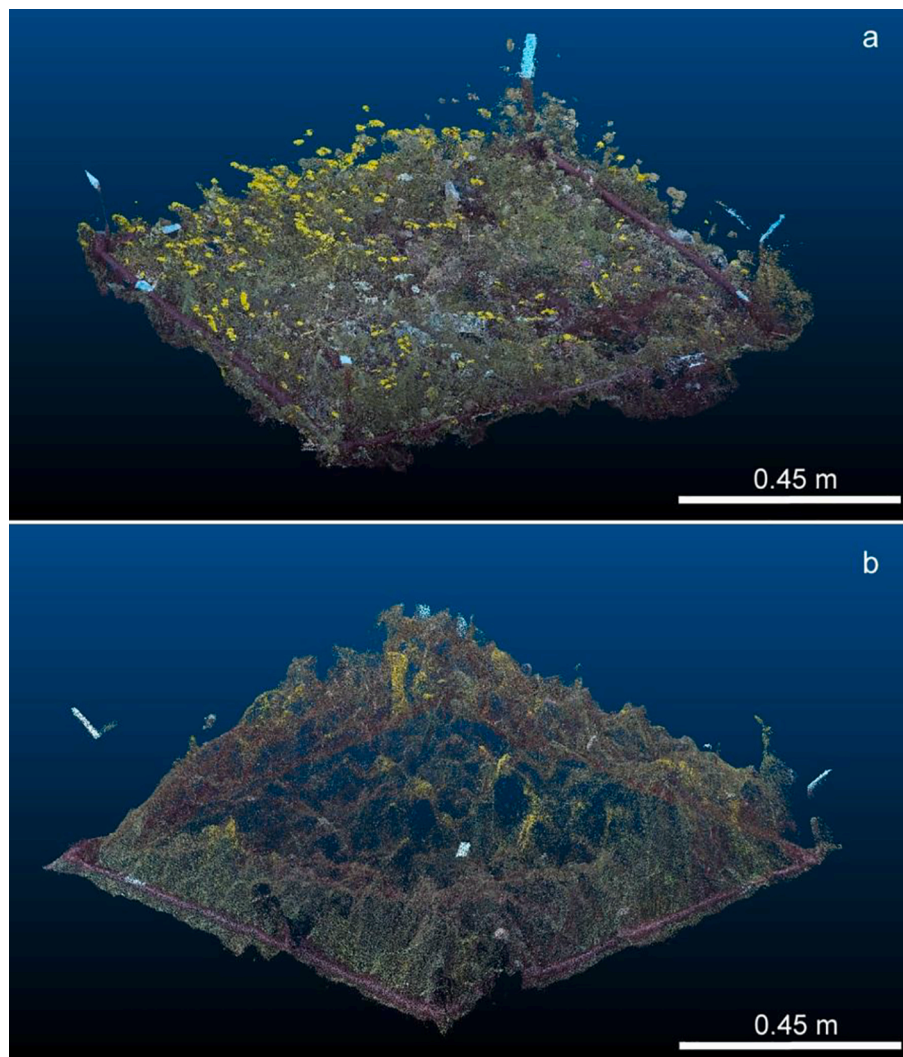


Fig. 3. Point cloud reconstruction of a xerophytic (a) and a mesophytic (b) plot.

Table 1

Statistics of fresh (FW) and dry (DW) weight biomass, and of canopy height (H), volume (V), volume adjusted (V_{adj}), and cover volume (V_{hc}): mean (μ), minimum (min) and maximum (max) values, and coefficient of variation (CV) are reported for all measurements and for mesophytic (MES) and xerophytic (XER) communities, and in different sites: Monte Serra (MS), Piano Ruggio (PR), and Serra Ciavole (SC). Units are indicated in brackets.

		μ	min	max	CV
FW (g)	Tot.	242.26	1.56	1672.65	162
	MES	381.41	13.20	1672.65	135
	XER	103.12	1.56	444.17	124
	MS	183.41	45.72	444.17	79
	PR	532.61	47.81	1672.65	107
	SC	10.80	1.56	24.87	89
DW (g)	Tot.	78.11	0.52	413.04	133
	MES	162.21	19.04	413.04	85
	XER	39.48	0.52	128.59	111
	MS	67.82	21.46	140.19	73
	PR	158.47	19.04	413.04	85
	SC	4.30	0.52	10.52	90
H (mm)	Tot.	53.28	2.57	216.99	98
	MES	70.66	9.36	216.99	94
	XER	35.91	2.57	80.72	69
	MS	49.70	24.06	80.73	42
	PR	98.64	38.06	217.00	66
	SC	11.54	2.57	19.45	51
V (dm ³)	Tot.	12.15	0.07	54.41	108
	MES	16.22	1.87	54.41	104
	XER	8.08	0.07	19.24	81
	MS	11.52	7.03	19.24	35
	PR	22.19	2.93	54.42	83
	SC	2.76	0.07	8.20	88
V_{adj} (dm ³)	Tot.	56.23	0.07	280.37	121
	MES	80.53	2.35	280.37	107
	XER	31.93	0.07	91.24	96
	MS	54.53	20.68	91.24	51
	PR	110.55	17.04	280.37	81
	SC	3.64	0.07	10.41	84
V_{hc} (dm ³)	Tot.	46.74	0.11	216.99	116
	MES	64.90	2.80	216.99	107
	XER	28.58	0.11	71.27	96
	MS	40.48	20.45	68.13	40
	PR	95.82	24.64	217.00	71
	SC	3.95	0.11	11.70	88

67.82 g, $P < 0.005$) (Table 1). Instead, no significant differences occurred between PR and MS ($P > 0.05$) (Table 1). Such productivity patterns among sites and communities were confirmed by the AGB predictors (Table 1).

3.2. Analysis of model performance

Overall performance and other statistical parameters indicated that all AGB predictive models attained a high accuracy (Table 2), with a coefficient of determination ranging from 90 to 96.5%. Furthermore, all models resulted more successful than FW in predicting DW (data not

Table 2

Relationships between dry weighted biomass (DW) and: canopy height (H), estimated volume (V), adjusted volume (V_{adj}) and volume from height and cover (V_{hc}). The first three columns compare the assumptions fitting of the models: influencers (maximum Cook distance, $D_{C_{max}}$), residuals normal distribution (significance of Shapiro-Wilk test, W), and residuals heteroscedasticity (Breusch-Pagan statistics, LM); the following three columns compare the overall performance of the model: predictivity (coefficient of determination, R^2), and distance from the model (root-mean-square deviation, RMSE; and relative root-mean-square deviation, rRMSE); two columns compare slope (a) and intercept (b) coefficients (value /standard error); (* = $P < 0.05$; ** = $P < 0.005$; *** = $P < 0.0005$). Leave-one-out cross validation is also reported (R^2_{cv} , and $RMSE_{cv}$).

	$D_{C_{max}}$	W	LM	R^2	RMSE	rRMSE	a	b	R^2_{cv}	$RMSE_{cv}$
H	0.23	>0.05	0.38	96.5	20.0	18.8	1.96***/0.09	-26.2***/5.8	96.0	10.3
V	0.38	>0.05	2.85	90.1	33.4	31.0	7.48***/0.52	-12.8/9.3	87.7	4.5
V_{adj}	0.32	>0.05	3.16	96.1	21.0	19.8	1.50***/0.06	-6.4/5.6	95.1	14.6
V_{hc}	0.40	>0.05	0.79	96.0	21.2	19.9	1.88***/0.08	-9.8/5.8	95.5	11.3

showed). However, the measured volume (V), albeit highly correlated to DW, resulted the less effective ($R^2 = 90.1\%$, $RMSE = 33.4$) among the tested AGB predictors. The other three predictors were highly comparable in their predictive power. The average canopy height (H) produced the model with the best fitting, and it was the only one with a significant intercept value (Table 2, Fig. 4a). Instead, the main differences between V_{adj} and V_{hc} regarded the model coefficients (Table 2, Fig. 4cd). Cross validation had no significant effect on coefficients of determination, which generally exceeded 95%. V instead showed a more marked a reduction of R^2 (Table 2). In all four models, the residuals were normally distributed and did not have significant correlation to DW.

The best non-linear fit of RW_{abs} on DW was represented by a power-inverse function (Fig. 5): therefore, when DW tends to zero, the RW_{abs} grows up abruptly, whereas going toward bigger DW, RW_{abs} tends to zero. In this model, the 10% error level was at a very low DW value (~9g). However, the measured data showed a meaningful deviation from the model at about 50 g, and lower DW values resulted in RW_{abs} over 0.3. Among the considered AGB predictors, V_{adj} resulted the best predictor by virtue of a lower RW_{abs} average value ($\mu = 1.3$; Figs. 6 and 7), whereas H had the highest RW_{abs} ($\mu = 3.5$); however, the differences among predictors were not statistically significant ($P > 0.05$). On average, all predictors marked a light DW overestimation (Fig. 8a); while, differences in RW average values were not significant ($P > 0.1$).

MES and XER did not show a significant difference in RW_{abs} average value ($P > 0.5$), evidencing that RW_{abs} was not related to the community type. Nevertheless, XER showed a higher RW_{abs} than MES, especially when using H ($\mu = 6.4$, and 0.6, respectively) (Fig. 6). On the contrary, V_{adj} revealed the lowest average difference ($\mu = 1.9$, and 0.6, respectively) (Fig. 6). Regarding DW over- and underestimation (RW, Fig. 8), XER was generally overestimated (μ ranging from 1.4, in V, and 4.1, in H; Fig. 8b), whereas MES was close to zero (μ ranging from -0.5, in V, and 0.3, in H; Fig. 8c).

Meaningful differences in RW and RW_{abs} occurred among sites, as SC possessed the largest RW_{abs} average value (Fig. 7) and the most variable RW (Fig. 8f). Even in this case, H resulted the predictor showing in SC the higher DW error level (RW_{abs} : $\mu = 9.5$) and overestimation (RW: $\mu = 7.3$).

4. Discussion

The studied communities have striking differences in terms of species composition, structure, cover and biomass (Gargano et al., 2017). Nonetheless, this study shows that the applied 3D-modelling approach can provide robust estimates of above ground biomass (ABG) in both the study contexts. Despite the marked differences within and between the sampled communities, the 3D models allowed for a very fine estimate of vegetation cover and canopy height, by virtue of a picking up score ranging from 195,000 to 878,000/m². Therefore, the measurements of the various AGB predictors considered in this study resulted to be extremely accurate in both communities. Hence, the relationships between such predictors and the direct AGB measurements were much stronger than those commonly found with other methods (rarely

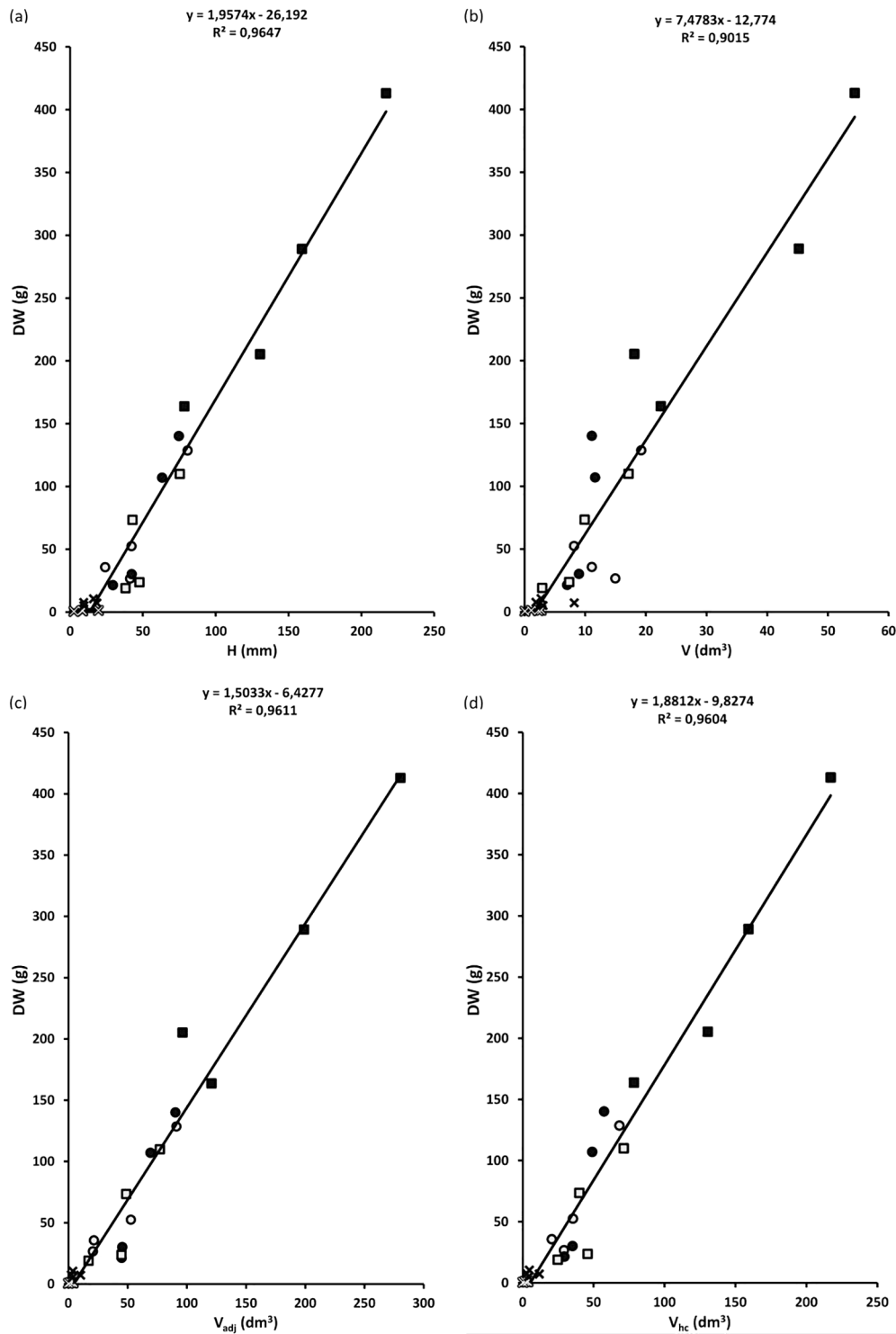


Fig. 4. Linear regression models of dry weight biomass (DW) on the four predictors: canopy height (H, a), volume (V, b), volume adjusted (V_{adj}, c), and cover volume (V_{hc}, d); the regression equation is reported over the relative diagram. Dark and light symbols refer to mesophytic (MES) and xerophytic (XER) communities, respectively. The sites are identified by the symbol shape: Piano Ruggio (PR, square), Monte Serra (MS, circle), and Serra Ciavole (SC, X).

reaching $R^2 = 96\%$).

Traditional approaches can be little effective in predicting biomass in wild grasslands because they consider just a few predictors (e.g., vegetation height), or due to the approximation in the measurements (e.g., visual cover estimation). Working with managed and cultivated pastures, such limitations can have a minor relevance. Indeed, being homogenous in structure and composition, such communities typically have a reduced spatial variability in vegetation cover and height. In

contrast, the high small-scale heterogeneity of wild grasslands can severely reduce the effectiveness of such simple methods in predicting AGB. This would suggest the use of more advanced techniques like 3D modelling. Accordingly, the comparison of biomass estimations based on volume and canopy height measures, revealed that 3D models are more effective than the plate meter approach in both wild grasslands (Cooper et al., 2017) and managed pastures (Wigley et al., 2019). Similar results were obtained by evaluating the AGB estimations

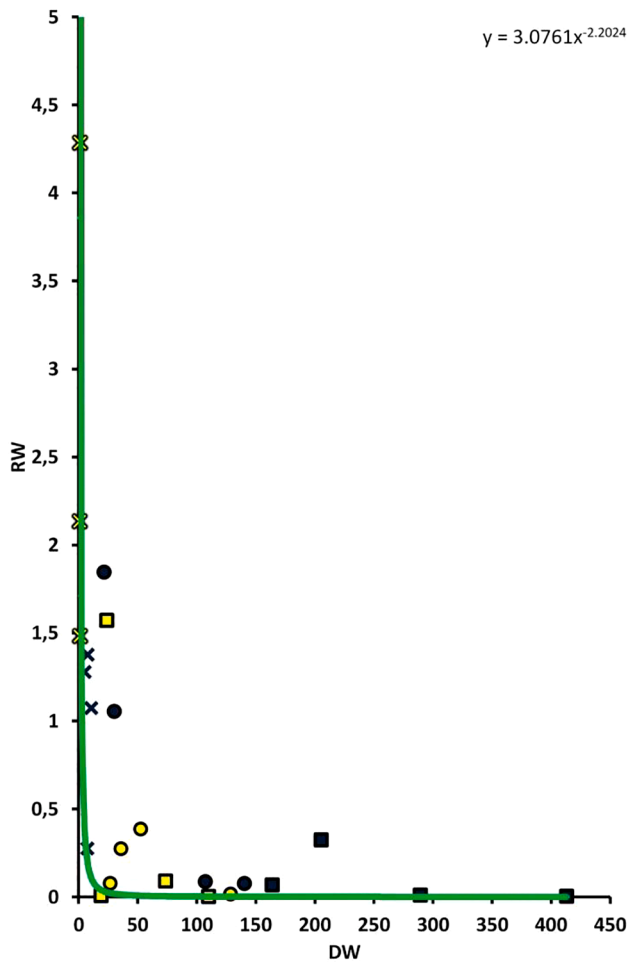


Fig. 5. Scatterplot of relative absolute weight (RW_{abs}) on dry weight biomass (DW) for V_{adj} predictor. Symbols are as in Fig. 1. The power-inverse function is over imposed.

obtained from 3D reconstructions of images provided by unmanned aerial vehicle (UAV). Indeed, also in such cases, the biomass predictions derived from 3D models revealed a better (Bareth and Schellberg, 2018; Lussem et al., 2019) or similar (Grüner et al., 2019) accuracy than those obtained from rising plate meter or ruler height measurements. The 3D reconstructions obtained from drone images can be useful for carrying out biomass estimations over large surfaces and combining them with spectroradiometer measurements. However, such estimates can be less accurate even if working on homogeneous herbaceous communities. A possible reason is that the resolution of the images acquired from UAV is lower than those obtained by working on a small plot at about 1 m distance. For example, Grüner et al. (2019) obtained a R^2 ranging from 58 to 78% with a pixel resolution between 7 and 8 mm; Lussem et al. (2019) reached a R^2 of 88% with a resolution between 1 and 2 cm. It is interesting to note that canopy height is the most used predictor, opening the opportunity of combining UAV and field 3D reconstructions.

Although the robustness of our models was confirmed in both study communities, some differences were found among sites. Overall, our results evidenced that a meaningful error level affected the biomass estimates carried out at SC (i.e. 10% error level at about 50 g/m^2 , instead of a theoretically expected value of 10% at 9 g/m^2). Such a discrepancy seemed a consequence of the very low AGB found in SC, which was caused by the intense grazing pressure generally affecting the high-mountain belt of the study area (Gargano et al., 2012). Accordingly, problems in obtaining precise biomass estimates were already found in heavily grazed pastures (Earle and McGowan, 1979; Murphy et al., 1995). In such contexts, it was argued that the disturbance caused by trampled biomass can be a primary error source for post-grazing AGB estimations (Stockdale and Kelly, 1984). Therefore, the appearance of such an error threshold could be interpreted as a signal of inadequate management (i.e. excessive grazing pressure).

The comparison of the predictive power of the four AGB proxies indicated that identifying the best predictor is challenging. Previous work demonstrated that measures of vegetation volume derived from 3D-reconstructions have a good correlation ($R^2 \sim 70\%$) to direct AGB measurements (Cooper et al., 2017; Passalacqua et al., 2019; Wallace et al., 2017). Instead, papers testing the effectiveness of canopy height as

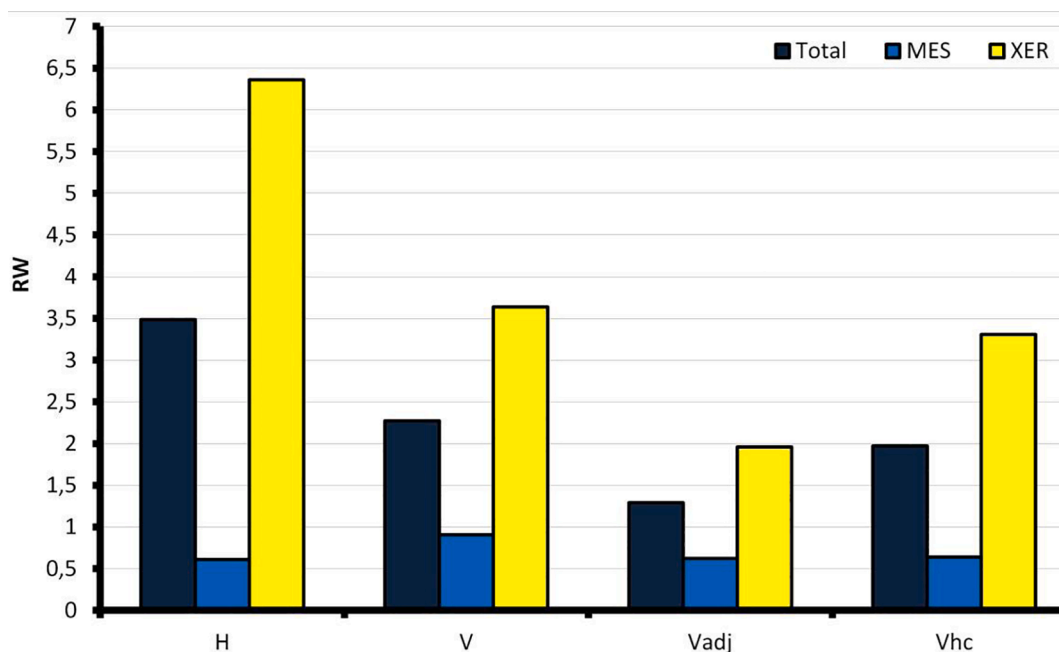


Fig. 6. Bar chart showing the average absolute relative weight (RW_{abs}) for four AGB predictors: canopy height (H), volume (V), volume adjusted (V_{adj}), and cover volume (V_{hc}), for all cases (Total), mesophytic communities (MES), and xerophytic communities (XER).

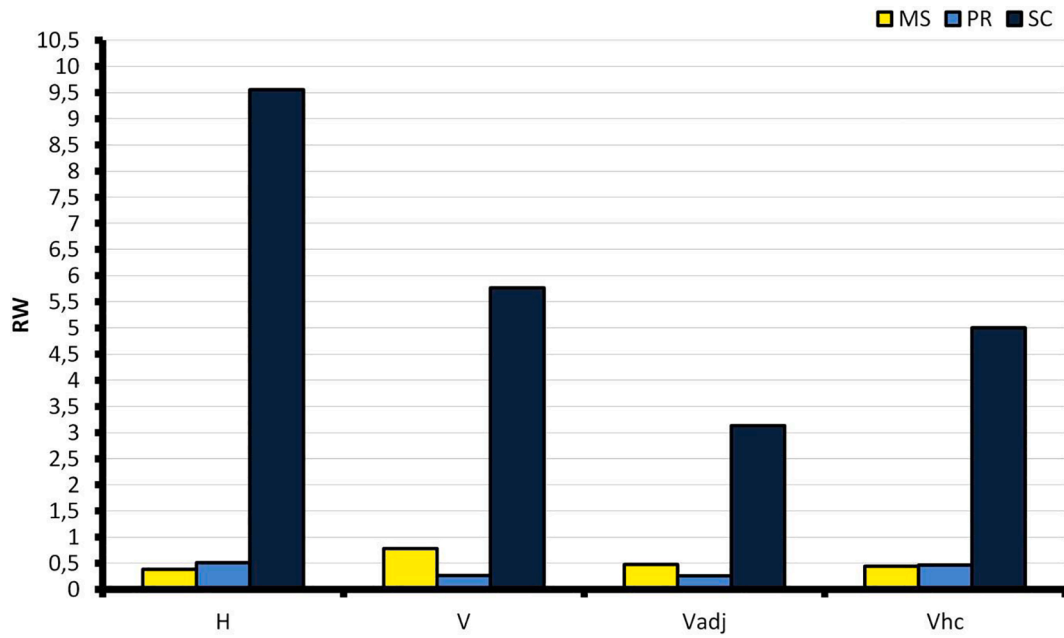


Fig. 7. Bar chart showing the average absolute relative weight (RW_{abs}) resulted for the three study sites based on four AGB predictors: canopy height (H), volume (V), volume adjusted (V_{adj}), and cover volume (V_{hc}) in Monte Serra (MS), Piano Ruggio (PR), and Serra Ciavole (SC).

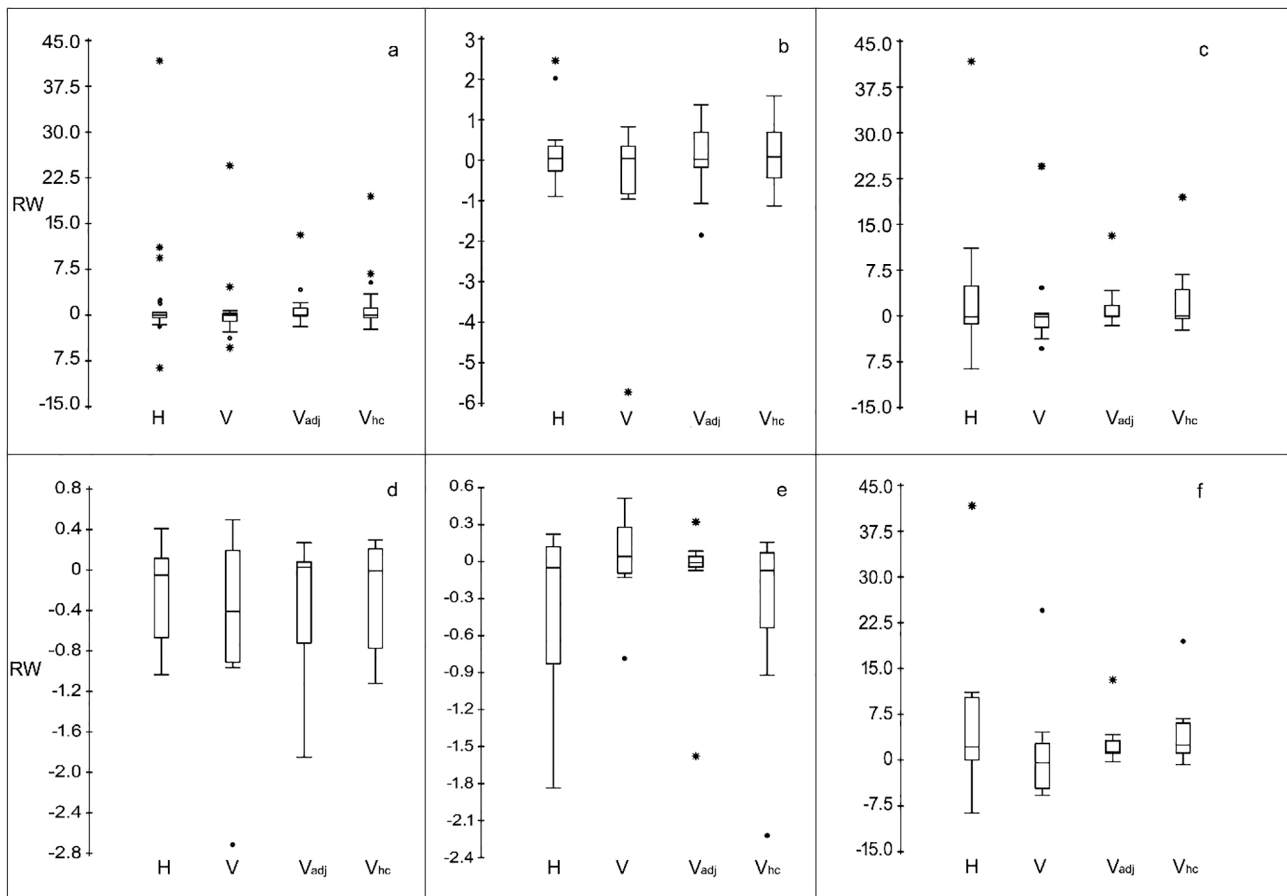


Fig. 8. Boxplots showing relative weight (RW) distribution based on t four AGB predictors: canopy height (H), volume (V), volume adjusted (V_{adj}), and cover volume (V_{hc}), for all cases (a), mesophytic communities (MES, b), xerophytic communities (XER, c) and in Monte Serra (MS, d), Piano Ruggio (PR, e), and Serra Ciavole (SC, f).

AGB proxy revealed correlation scores from good (Grüner et al., 2019; Hütt et al., 2014; Lussem et al., 2019; Schulze-Brüninghoff et al., 2019), to high (Bareth and Schellberg, 2018; Wigley et al., 2019). Analogously, comparing four AGB predictors derived from TLS-3D point clouds, Schulze found that predictors based on measures of canopy height have a better fit than volume-related proxies. However, the correlations found by Schulze-Brüninghoff et al. (2019) were only moderate to good; moreover, the tested predictors were more related to fresh biomass rather than dry one.

Substantially, our work confirmed the average canopy height as the best biomass predictor. In our models, it was also the only predictor with a significant intercept coefficient, which is a further indication of model reliability. From a functional viewpoint, plant height is typically retained to be an indicator of competitive vigour, which can be related to individual reproductive success, and tolerance versus environmental stress (Cornelissen et al., 2003). For such reasons, plant height is considered a meaningful trait to be measured in plant monitoring programs (de Bello et al., 2010). Therefore, albeit further investigations are required, our findings highlight that the average canopy height is a relevant trait at higher ecological levels too, because it is a robust predictor of above ground biomass, which in turn represents a major indicator of ecosystem functioning. In addition, our models indicated that canopy height-derived AGB estimations are little affected by variations of cloud point density and vegetation spatial heterogeneity; this would limit the risk of sampling and computational errors.

With regards to the other tested AGB proxies, the volume calculated by the raster grid (V) resulted the least accurate in predicting AGB. In particular, it was affected by a higher error related to the 3D reconstruction quality. This is a crucial point when applying the SfM approach, because the quality of point clouds depends on several factors linked to the operator ability and the environmental conditions in which the photograms are acquired. Indeed, according to previous work (Passalacqua et al., 2019), adjusting the volume by the coefficient of density (V_{adj}) significantly improved the predictive power. Instead, V_{adj} and V_{hc} showed a similar accuracy in predicting biomass: however, the V_{hc} model had a slightly lower fit than the V_{adj} one. Our findings suggest that adjusting the volume by the coefficient of density is the best option for using volume data derived from 3D point clouds in order to estimate AGB.

By comparing H and V_{adj} , the former resulted to have a higher correlation to actual biomass, but its variation range was narrower than observed in V_{adj} . Usually, variables with a greater variability are expected to have a larger effect on the dependent variables (Vázquez et al., 2005). Accordingly, Grinath (2019) found a clear relationship between predictor variability and its ability to predict biomass, suggesting that biomass is better estimated by predictors with relatively greater variability. Apparently, our findings did not fit such a relationships. However, looking at the intercept coefficient, the V_{adj} model revealed a better fit than that the H one (i.e. the intercept value closer to 0). Likely, this was the consequence of a higher power to estimate biomass at lower DW values. Indeed, the average RW_{abs} was lower in V_{adj} than in H , and this would have contributed to reduce the intercept value close to zero.

Our results offer some promising indications for defining a standardized protocol for monitoring AGB in herbaceous communities. In the first step, such an approach would require biomass harvesting from several 1 m^2 vegetation plots to investigate the relationships between direct biomass measures and predictors derived from 3D point clouds. The best relationship can be then applied to estimate biomass, without vegetation harvesting, in further plots, which could be used as reference units in long-term monitoring programs. In addition, the applied fine-scale approach could be combined with other remote sensing methods (i.e. SfMs from UAV) to improve the monitoring potential. For instance, the values of vegetation volume/canopy height determined from the fixed plots could be used as a reference to calibrate 3D reconstructions from UAVs. This would enhance the accuracy of biomass estimates over larger habitat surfaces.

5. Conclusions

Overall, our models had a high fit under all the study conditions. This confirms that AGB proxies computed from 3D vegetation reconstructions can be an effective way to evaluate primary productivity in herbaceous communities with complex structure and composition patterns. As a major limitation for this approach, we detected some loss of predictivity power at very low productivity rates. Therefore, the contexts characterized by a very poor vegetation cover (i.e. communities inhabiting extreme environments, heavily grazed and trampled pastures) may challenge the reliability of this method. Probably, in such conditions, traditional AGB measures would be preferable.

The canopy average height (H) deserves consideration when this kind of analyses are performed, because resulted the most performing predictor showing the best R^2 and RMSE; besides, it can be used to calibrate biomass estimates performed with 3D reconstructions made by the use of UAV. However, the vegetation volume, once adjusted by the coefficient of density, resulted to be the most effective proxy for predicting true values of above ground biomass, showing the lowest error in biomass estimation. Therefore, values of V_{adj} obtained by 3D vegetation models could be routinely used as an effective and non-destructive indicator for monitoring the trends of ecosystem productivity and functioning in habitats of particular conservation concern.

CRedit authorship contribution statement

Nicodemo G. Passalacqua: Conceptualization, Methodology, Formal analysis, Investigation, Writing - original draft, Writing - review & editing, Visualization, Supervision. **Simona Aiello:** Software, Formal analysis, Data Curation. **Liliana Bernardo:** Investigation, Resources. **Domenico Gargano:** Investigation, Writing - review & editing, Funding acquisition.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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