

## Article

# An Assessment of Proso Millet as an Alternative Summer Cereal Crop in the Mediterranean Basin

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**Abstract:** Proso millet (*Panicum miliaceum* L.) is a cereal well known for its ability to be successfully grown under drought and intense heat conditions, thus sustaining food security in arid regions. Considering that a trend of increasing drought severity is expected in the future in Southern Europe, solutions need to be found to enhance the resilience of agroecosystems to the effects of climate change. From this perspective, proso millet re-introduction could represent an interesting tool in reducing water consumption for grain production and in providing a new resource to farmers. The aim of this study was to characterize proso millet adaptability to drought and low-input field conditions in the Mediterranean environment, especially considering water-related traits, such as water use efficiency. Limited water-demanding crops and yield stability can contribute to the resilience of agroecosystems and their adaptation to climate change. A three-year field crop experiment was conducted in northern Italy to assess proso millet's performance in terms of productivity and water status in rainfed agriculture conditions. It was compared to a conventional irrigated corn, a typical summer cereal of the area. All years of experimentation were characterized by adverse meteorological trends, in the full manifestation of the uncertainties of climate change. Despite such different conditions from an agro-meteorological point of view, proso millet showed, in non-irrigated conditions, stable yield and water use efficiency (on average 0.30 kg/m<sup>2</sup> and 1.83 kg/m<sup>3</sup>, respectively), and good agronomic performance. Proso millet, therefore, seems to offer interesting traits for reintroduction on the European side of the Mediterranean Basin, representing a resource for farmers. Moreover, the shortness of the proso millet life cycle (on average 108 days) allows it to be used as a catch crop in the event of major crop failure, an event becoming more likely in the climate change scenario. Furthermore, the possibility of producing grain while saving water (and other production inputs), even in very hot and dry years, increases the sustainability of agricultural production and the resilience of agroecosystems.

**Keywords:** proso millet; climate change; resilience; dry land cereals

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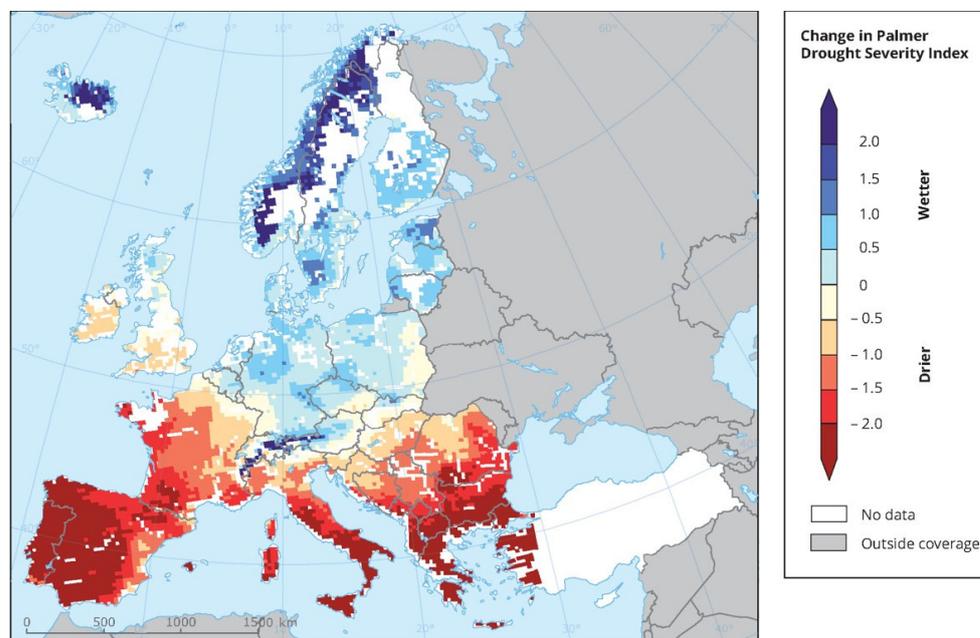


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## 1. Introduction

In the area of the Mediterranean Basin, climate change is characterized by high temperature with frequent heat waves, erratic precipitation, and extreme meteorological events (both floods and droughts). In particular, a trend of increasing drought severity is expected to show up in the future, as reported by [1]. The Palmer Drought Severity Index (PDSI) is a standardized index of drought that uses precipitation and temperature data to measure the cumulative deficit (relative to local mean conditions) in surface land

moisture. Figure 1 shows the projected change of PDSI in the 2021–2050 period compared to the thirty-year period from 1961–1990 in Europe. This scenario of increasing heat stress occurrence and longer dry spells poses a serious threat to summer crop yield, as well as to the quality of crop production, also making a proper crop choice for farmers difficult [2].



**Figure 1.** Projected PDSI change between 1961–1990 and 2021–2050 using 12 regional climate models (RCMs). Red and blue, respectively, indicate drier and wetter conditions. Source: European Environmental Agency, Indicator assessment (2017). <https://www.eea.europa.eu/data-and-maps/indicators/water-retention-4/assessment> (accessed on 30 January 2022).

Proso millet (*Panicum miliaceum* L.) could have great potential to be adopted as a promising crop resilient to climate change in the Mediterranean Basin. Millet used to be cultivated in ancient times in the Po Plain area. In particular, proso millet was the main species cultivated in the north of Italy after the Bronze age [3], and it was abandoned in favor of maize later on. It is now necessary to redefine agricultural practices and management and describe its phenology. It is an annual herbaceous plant of the Gramineae family. It has optimal nutrition parameters in terms of protein, minerals, and micronutrient content [4]. Millet is the sixth world's most important cereal, sustaining food security in arid regions and marginal lands [4]. In developed countries, as a low-demanding additional source of income, it can be grown as a secondary crop with winter cereals or as a catch crop in case of major crop failure [5,6]. Proso millet can be successfully grown under drought and intense heat conditions in arid non-irrigated lands with just 200–500 mm of average annual precipitation [7,8]. In fact, as reported by [9], proso millet has a shallow root system that is generally limited to the first 90 cm of soil and is really efficient at removing water from the topsoil and converting it into grain. Thus, it requires very little water compared to other cereals and can survive on topsoil moisture and summer precipitation, without the need for irrigation, restoring subsoil moisture for subsequent crops with a deeper root system. It also avoids drought sensitivity by having a really short cycle, carried out in 60–90 days [7]. In addition, proso millet is a C4 crop whose advantages in terms of drought stress resistance are well known [10,11]. Moreover, it can successfully adapt to various pedological conditions, such as saline, low fertility, and slightly acid soils [4]. As a warm season grass, proso millet needs to be planted in spring (soil temperature requested for a good germination must be at least 12 °C, as reported in [7]) in a moist and firm seedbed. The first two weeks after planting are

considered the most critical time in proso millet cultivation. This is due to the fact that final yield highly depends on soil water content at planting, so light precipitation is very helpful, but heavy rain can have a negative effect [9]. Generally and in particular after a fallow year, proso millet does not need any fertilization. The effects of drought stress on yield and water use efficiency (WUE) clearly depend on the phenological stage in which stress appears and on its severity. Water stress at the ear emergence stage causes the greatest grain yield loss (around 40%) due to the reduction of both seed number per ear (as a result of stress on pollination and floret abortion) and seed weight (as a result of cytokinin reduction, which causes less endosperm production) [12–14]. As reported in [15], the seed filling stage appears to be less susceptible to drought stress. The proso millet panicle shows a staggered ripening that starts from the top and gradually continues to the bottom. Generally, as the bottom part of the panicle reaches full-ripening, the distal part of it starts grain loosening. Grain shattering causes yield loss if harvest is delayed [9,16]. At maturity, grains generally present about 20% or less moisture.

This case study of proso millet cultivation in the Mediterranean Basin is in the frame of a LIFE-CCA EU project, called Growing REsilience AgriculTure (GREAT LIFE). The goal of GREAT LIFE is to face the effects of climate change on agricultural activities in Italy and on the European side of the Mediterranean Basin in general. Through the experimentation of rational rotation schemes and sustainable agronomic practices, the project aims to experiment with stress-resistant low-demanding crops as potential alternatives to maize in crop rotation at the Italian and European levels to improve the resilience of agroecosystems and reduce water consumption. In fact, in addition to being an extremely water demanding crop (thus reducing its sustainability), corn productivity is becoming less reliable and profitable for farmers in Southern Europe as a consequence of climate change effects on corn yield [17]. From 1974 to 2008, the corn yield losses caused by climate change in Western and Southern Europe regions have been estimated by [17] to be around 6.3%. In addition, climate change impacts are not just related to quantitative crop performances but also to quality aspects, favoring the increase of mycotoxins and corn pathogens. In fact, a recent study focusing on aflatoxin contamination in maize within the next 100 years, under a +2 °C and +5 °C climate change scenario, demonstrated that the increasing temperatures clearly are closely related to aflatoxin contamination risk [18]. The exact definition of proso millet phenology encoded in the BBCH (Biologische Bundesanstalt, Bundessortenamt and CHEmical industry) scale is an important part of the GREAT LIFE project since it allows progress in research and gives researchers comprehensive indications for future agronomic surveys on the crop. Additionally, the calculation of temperature-driven heat-unit accumulation (cumulative growing degree days, CGDD) is important, and knowing the relationship between CGDD, days after sowing (DAS), and phenological BBCH stages can help to successfully cultivate proso millet, allowing us to identify the best sowing time and the most susceptible phases to abiotic stress during the life cycle. For this reason, in the first year of experimentation, *P. miliaceum* L. phenological development was encoded in the BBCH scale, also indicating the thresholds in terms of CGDD and DAS necessary to achieve each phase (for more details, refer to [19]). The aim of this study was to assess proso millet's ability to maintain a high vegetative vigor and a good water status (comparable to that of an irrigated corn) without any irrigation, as well as to evaluate its resilience to climate change conditions in the Mediterranean climate.

## 2. Materials and Methods

### 2.1. Experimentation

Through a three-year (2019, 2020, 2021) open field experiment, proso millet was cultivated in the Emilia-Romagna region, without irrigation and cultivation treatments (fertilizations or others). The aim was to gather as much information as possible on the possibilities offered by proso millet as a promising low-demanding, drought-resistant

crop in improving the resilience of agroecosystems to climate change, reducing the consumption of water and other resources for grain production, and providing a new resource to farmers. During the field trial, agronomical parameters were collected on the crop, including phenological, physiological, and biological data. Proso millet agronomic performance (in terms of grain yield and water use efficiency) and vegetation indexes were compared with those of irrigated corn cropped in an open field in a conventional productive context.

## 2.2. Experimental Sites

The site chosen for the proso millet open field test in 2019, 2020, and 2021 was in the Emilia–Romagna region, at the Azienda Villa Masini (VM) organic farm (Ravenna, Lat 44°15'59", Long 12°07'48"). Here, the experimental field has a size of one ha.

As regards maize, yield and water balance data were collected in an experimental field located at Aqua Campus (AC), in the Municipality of Budrio (Bologna, Lat 44°33'46" Long 11°32'23"); the AC experimental field size was 0.4 ha. Aqua Campus is an experimental site run by CER (Consorzio per il Canale Emiliano Romagnolo-<https://consorzio-cer.it/it/> - accessed on 16 December 2021), with the purpose of developing smart irrigation practices to make water use in agriculture more and more efficient. Therefore, AC is not considered an experimental site of the GREAT LIFE project, but AC data were used as a reference for irrigated corn in the area to compare the performance of proso millet in our experiment with the current standard grain summer crop in the region.

Moreover, we used phenological data for comparison in 2019 and 2020 agronomic seasons at a second site, the Agricultural Garden (AG) of the Department of Agricultural and Food Sciences, University of Bologna (DISTAL), inside the Department Campus in Bologna (Lat 44°30'54" Long 11°24'21"). Here, plots have a size of 2 × 2 m<sup>2</sup>, and millet was introduced in the normal Agricultural Garden cultivation scheme. This plot had the aim of giving the possibility to strictly follow the phenological development of plants. In 2021, reliable data could not be obtained from the AG site due to a strong delay in crop establishment after planting. For this reason, the comparison of the thermal thresholds relates only to the first two years of experimentation.

## 2.3. Agronomic Management

Agronomic management for all years and for the three experimental sites, with planting and harvest day, is presented in Table 1. At the VM experimental site, soil analyses were carried out each year before sowing, considering principal physical and chemical properties, and are reported in Table 2.

**Table 1.** Agronomic management for the three experimental sites.

	AG	VM	AC
Crop	Proso millet ("Miglio Biondo"- B109-SFU-from Arcoiris company)	Proso millet ("Miglio Biondo"-B109-SFU-from Arcoiris company)	Maize ("Sistematico", from Società Italiana Sementi)
Soil management	Use of cultivator at the end of previous season	2 false seedbeds before sowing	Ploughing, grubbing, harrowing
Planting date	18 April 2019 27 April 2020	17 April 2019 27 April 2020 22 April 2021	20 March 2019 26 March 2020 24 March 2021
Sowing depth	2 cm	2 cm	3–4 cm
Inter-row	8 cm	8 cm	70 cm
Seed density	4 (g/m <sup>2</sup> )	4 (g/m <sup>2</sup> )	7 plant/m <sup>2</sup>

Fertilization	none	none	Universal UP (350 kg/ha) Nitroslow (275 kg/ha) Ammonium Nitrate 27% (100 kg/ha) Nitroslow (175 kg/ha)
Irrigation	none	none	180 mm (2019) 190 mm (2020) 213 mm (2021)
Weeding method	manual	none	Both chemical and mechanical
Harvest date	07 August 2019 20 July 2020	01 August 2019 12 August 2020 13 August 2021	29 August 2019 02 September 2020 01 September 2021

**Table 2.** Physical and chemical soil characteristics at the VM experimental site.

Year	2019	2020	2021
pH	7.74	8.11	7.82
Organic matter (g/kg)	17	16	14
C/N ratio	9.1	10.5	10
Total N (g/kg)	1.1	0.6	0.8
NH <sub>4</sub> <sup>+</sup> -N (g/kg)	<0.002	<0.002	<0.002
NO <sub>3</sub> <sup>-</sup> -N mg/kg	12	39	33
P <sub>2</sub> O <sub>5</sub> <sup>-</sup> mg/kg dm	<9	11	15
K <sub>2</sub> O mg/kg dm	170	235	302
<b>Granulometry</b>			
Clay (%)		24	
Silt (%)		58	
Sand (%)		18	

## 2.4. Data Collection

### 2.4.1. Meteorological Data and Water Balance

Agrometeorological data (maximum, minimum, average daily air temperature, and daily precipitation) were monitored throughout 2019 and 2020 cropping seasons for the three experimental sites and for VM and AC sites in 2021. For AG, data were obtained from the DISTAL agrometeorological station present in the same site, for VM from the ARPAE-Simc agrometeorological station of San Pietro in Vincoli, which is closest to the experimental field (approximately 4 km far). For part of the 2021 season, precipitation data were collected from the ARPAE-Simc agrometeorological station of Cocolia (also approximately 4 km far from the VM site), due to missing data from San Pietro in Vincoli. For maize, agrometeorological data were provided by the registration of the plot on the IRRIFRAME portal (<https://www.irriframe.it/Irriframe> - accessed on 16 December 2021, service offered by ANBI - Associazione Nazionale Consorzi di gestione e tutela del territorio e acque irrigue), coordinated by CER (Consorzio per il Canale Emiliano Romagnolo). The IRRIFRAME service was also used to gather the calculations of crop water balance for corn at the AC site. IRRIFRAME services are Irrigation Advisory Services for Farm Water Management based on a water balance model aimed at crop irrigation management at a field scale. The model structure takes into account the soil–plant–atmosphere continuum,

including soil water balance, plant development and atmospheric thermal regime, rainfall, and evaporative demand ([https://www.irriframe.it/irriframe/Content/IrriFrame\\_Documentation\\_english\\_version.pdf](https://www.irriframe.it/irriframe/Content/IrriFrame_Documentation_english_version.pdf) - accessed on 6 December 2021).

If millet crop parameters were not available on the IRRIFRAME PORTAL, the calculation of millet water balance at the VM site was performed as follows:

$$ETc \text{ (crop seasonal evapotranspiration)} = P + \Delta\theta + I + UP - \text{RUN} - \text{PERC}$$

Where:

P = precipitation;

$\Delta\theta$  = variation of soil moisture from start to end of crop cycle in the root zone;

I = irrigation;

UP = capillary rise from the groundwater table in the root zone;

RUN = superficial run off;

PERC = percolation in the deep soil layers.

All terms for water balance are expressed in mm. The P value was obtained from the ARPAE-Simc agrometeorological station sited in San Pietro in Vincoli, summing all precipitation events in the growing season.  $\Delta\theta$  was obtained from soil samples taken from the field at the beginning and end of the cycle at a depth of 30 cm, from the weight difference between fresh weight and dry weight (post drying in oven at 105°C for 24 h, conventional drying method [20]), and transformed in mm of water in the root zone. UP was calculated through UPFLOW software [21] on a daily basis, and then summed for the whole cycle. I was equal to zero, as stated in Table 1, and RUN and PERC were considered equal to zero, considering the pedological characteristics and the rainfall pattern in VM. Considering the very low contribution given by millet to evapotranspiration in the first stages of leaf development, we decided to start calculating ETc when BBCH stage 16 (six leaves fully expanded) was reached. Before BBCH stage 16, when crop ground coverage and biomass were highly restricted, evaporation from bare and nearly bare soil was quantified following the FAO method as described in FAO Irrigation and Drainage Paper 56 [22]. Water balance was calculated in order to relate yield to water consumption (water use efficiency, WUE).

#### 2.4.2. Phenological Data

Proso millet phenology was strictly monitored in both VM and AG sites in 2019 and 2020. The 2019 data collection was useful to encode proso millet phenology in the BBCH scale, including CGDD and DAS thresholds for reaching the various phases [19]. The 2020 data collection was useful to compare with the 2019 season in order to test the consistency of thermal thresholds identified in 2019. Maximum and minimum air temperature values were used to calculate CGDD from the sowing date with a temperature threshold for millet of 10 °C [23]. CGDD was calculated using the single triangle method [24,25], which gives good accuracy with an acceptable error level compared to hourly air temperature values [24,26]. Obviously, phenology monitoring in 2021 at the VM site was performed to relate crop vigor and resistance to water stress with the stages of the life cycle. Corn phenological development monitoring was offered by the IRRIFRAME service.

#### 2.4.3. Satellite Indexes

To monitor crop vegetative and water status, vegetative indexes obtained by processing satellite images downloaded from SENTINEL 2 were used, through the free ESA elaboration software SNAP. In relation to the European Copernicus environmental monitoring program, the European Space Agency (ESA) launched two identical satellites (SENTINEL-2A in 2015 and SENTINEL-2B in 2017) that operate simultaneously, phased at 180° to each other in a sun-synchronous orbit at a mean altitude of 786 km to ensure a high frequency of overflights and continuous availability (<https://senti->

nel.esa.int/web/sentinel/missions/sentinel-2/satellite-description - accessed on 6<sup>th</sup> December 2021). The two satellites are equipped with a Multi-Spectral Instrument (MSI) that allows the acquisition of multispectral images in 13 bands of the visible and infrared, with a resolution of ground details of 10, 20, and 60 m depending on the spectral band (<https://sentinel.esa.int/web/sentinel/missions/sentinel-2/overview> - accessed on 6<sup>th</sup> December 2021). With its frequent and systematic coverage, SENTINEL-2 represents a significant contribution to land monitoring services, providing data and supporting the assessment of biogeophysical parameters, such as leaf area index (LAI), leaf chlorophyll content (LCC), and leaf cover (LC) (<https://sentinel.esa.int/web/sentinel/missions/sentinel-2/thematic-areas-and-services/land-monitoring> - accessed on 7<sup>th</sup> December 2021). SENTINEL 2, therefore, allows significant uses in remote sensing applied to precision agriculture, allowing monitoring of the vegetative and health state of crops, thus representing an instrument in the decision support system. In this case study, we focused on two vegetation indexes gained from Sentinel MSI: NDVI and NDWI.

NDVI (normalized difference vegetation index) is the most widely used vegetation index that provides effective information on the vigor status of the crop. It is calculated as the ratio between the difference and the sum of the radiation reflected in Near InfraRed (NIR) and red (RED), ranging from -1 to +1:  $NDVI = (NIR - RED) / (NIR + RED)$  [27]. Values between -1 and 0 indicate anthropic areas or streams, and values > 0 are typical of areas covered by vegetation. Values closer to 1 indicate a crop with high field coverage and vigor. Usually, values higher than 0.7 indicate a crop with high soil cover and high vigor.

NDWI (normalized difference water index) is calculated as the ratio between the difference and the sum of the radiation reflected in Near InfraRed (NIR) and Short Wave InfraRed (SWIR), ranging from -1 to +1:  $NDWI = (NIR - SWIR) / (NIR + SWIR)$  [28]. Values between -1 and -0.8 indicate bare soil. Higher values indicate vegetation presence. The closer the values are to 1, the higher the vegetation cover and the better the water status of the crop (absence of stress). Values between 0.2 and 0.4 already indicate a crop with high soil cover and an absence of water stress. NDVI and NDWI were calculated for millet and corn in VM and AC sites, respectively, on three dates along the crop cycle, to monitor vigor and water status. These dates were determined by the satellite passage over the granule of interest (which included both experimental sites) and the contextual absence of cloud cover. Dates for the three years and crop phenological stages are reported in Table 3.

#### 2.4.4. SPAD

The SPAD (soil and plant analysis development) value was measured on millet during the three seasons and throughout the life cycle at the VM site (taken each time on 8 plants, each plant value is given by the average of 3 separate measures on the same leaf), as an indirect indicator of chlorophyll content. SPAD measurements were performed using the Konica Minolta SPAD-502 Plus Chlorophyll Meter (Konica Minolta Inc., Osaka, Japan).

#### 2.4.5. Agronomic Performance

To assess the agronomic performance of proso millet at the VM site, before harvesting, 3 separate sampling areas of 1 m<sup>2</sup> were evaluated for the following parameters: ground cover (%), weed presence on the monitored surface (%), yield per panicle (measured on 5 panicles), plant height (measured on 5 plants at the panicle insertion), and panicle length (measured on 5 panicles). Ground coverage and weed presence were both assessed through visual analysis of the soil surface. The percentage of ground cover and weed presence represent the ratio of crop vegetation and weed presence, respectively, with bare soil (with 0% = bare soil and 100% = soil not visible due to complete plant coverage). Then, the same 3 separated areas were threshed in order to quantify grain yield and compare it to water consumption for the determination of WUE.

CER kindly provided corn grain yield data from AC. Here, sampling areas of 10 m<sup>2</sup> were threshed in the 3 years (4, 2, and 3 replicates in 2019, 2020, and 2021, respectively). For both sites, grain yield was expressed as grain kg m<sup>-2</sup>, at 0% humidity (determined after drying in an oven at 105 °C for 72 h [29]).

### 2.5. Statistical Analysis

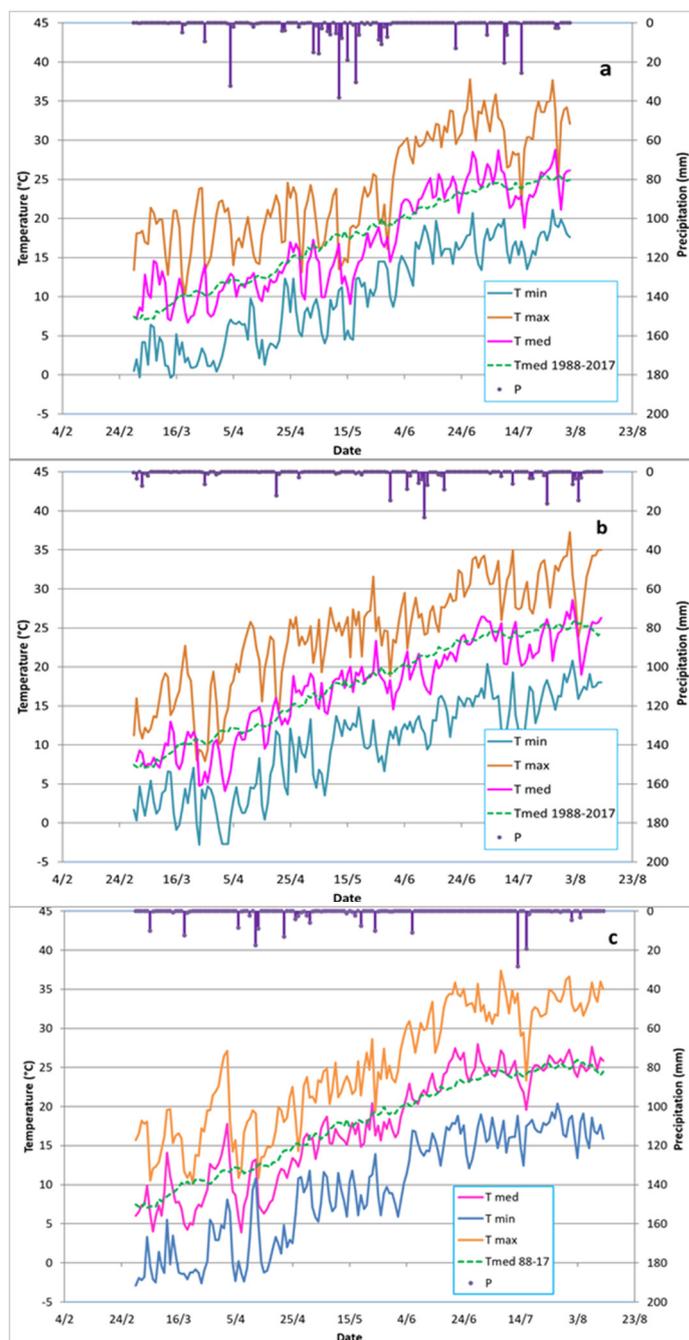
Statistical analysis on yield/panicle, plant height, and panicle length in the three years was performed using R software version 4.0.3. Data normal distribution and homoscedasticity, necessary assumptions to perform ANOVA, were verified for each trait with Shapiro–Wilk and Levene tests, respectively ( $p = 0.05$ ). When data did not meet the normal distribution assumption for ANOVA, they were transformed via logarithmic transformation to solve the problem, and Tukey’s post-hoc test was used to separate the retro-transformed means ( $p = 0.05$ ). When data did not meet the normal distribution assumption but were homoscedastic and data transformation was not a viable solution, a non-parametric Kruskal–Wallis test was performed. When data were normally distributed but not homoscedastic, the non-parametric Welch test was performed, and the Duncan–Waller test was used to separate the means, if the Welch test revealed significant differences ( $p = 0.05$ ).

## 3. Results

### 3.1. Meteorological Data

Figure 2a–c show the meteorological data for the VM site in 2019, 2020, and 2021, when proso millet was grown in an open field. Data from 2019, 2020, and 2021 were compared with the 30-year mean air temperature in Cadriano (Lat 44°33′03″, Lon 11°24′36″), where the main DISTAL agrometeorological station is located, for which a solid historical dataset is available. We also calculated the 30-year mean precipitation for an average crop cycle. The chosen period was 15 April to 15 August (cfr with sowing/harvesting data in Table 1). Climatological precipitation during the crop cycle was 230.8 mm, while for 2019 it amounted to 310.6 mm, quite a large quantity, and the two following years showed low rainfall quantities, namely 159.8 mm (in 2020) and 119.6 mm (in 2021). As it was more than the total quantity during the cycle, its distribution in single years is interesting.

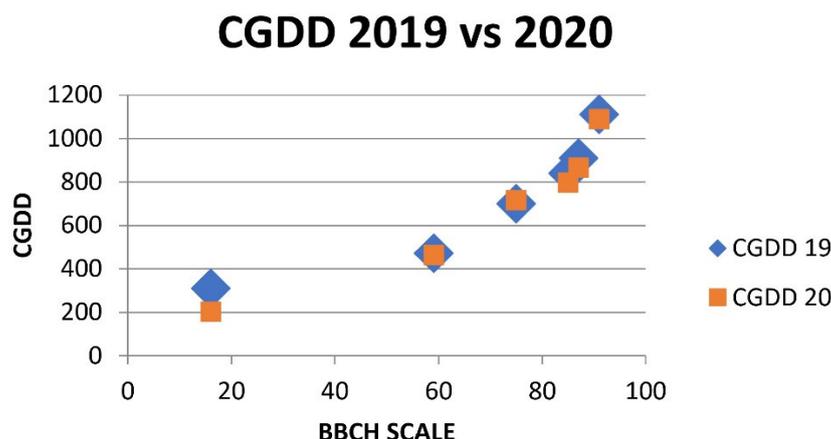
The 2019 season was characterized by a cold and rainy May (T mean 14.7 °C, most of the time under the climatic average, 183.6 mm of precipitation) during the BBCH stage 00–16 (from sowing to six fully expanded leaves), followed by a hot summer, with heat waves in June and July. In 2020, spring temperatures were mostly close to the climate, but precipitation was extremely restrained, with just 18.8 mm during the entire month of May, during BBCH stage 00–16. In 2021, only one precipitation event (11 mm) was recorded in the period, including the entire month of June and the first ten days of July. Two heat waves occurred in the same period, with temperature peaks over 35 °C (37.4 °C on July 7<sup>th</sup>), between the end of leaf development (BBCH 16) and flowering (BBCH 60–69). Although very different, 2019, 2020, and 2021 were three difficult seasons from an agronomical point of view, in the full manifestation of the uncertainties related to climate change.



**Figure 2.** Meteorological data for VM site in 2019 (a), 2020 (b), and 2021 (c). As reference, the 30 year mean air temperature (1988–2017) measured at the Cadriano station was added.

### 3.2. Phenological Development

Phenological data collected at the VM and AG sites in 2019 and 2020 have been compared to test the consistency of CGDD thresholds necessary to reach each stage. Figure 3 reports phenological data. The difference in the thermal thresholds between 2019 and 2020, expressed as the average between the 2 experimental sites, is mostly reduced, suggesting good reliability.



**Figure 3.** The CGDD necessary to reach the same BBCH stage in the 2019 and 2020 seasons.

### 3.3. Satellite Indexes

Table 3 reports NDVI and NDWI calculated for corn at the AC site and millet at the VM site during the 2019, 2020, and 2021 seasons. For both crops, the BBCH principal growth stage is reported at each date. As an example, Figure 4 shows vegetation indexes calculated with ESA SNAP software on 5 July 2019.

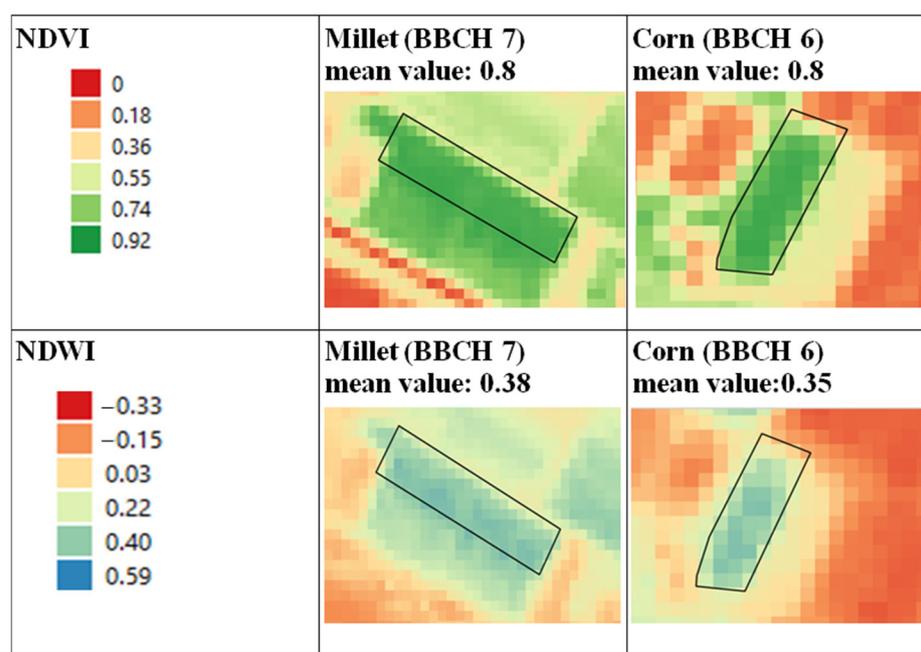
**Table 3.** Corn (in AC site) and millet (in the VM site) NDVI and NDWI in 2019, 2020, and 2021 seasons.

2019	18 June	05 July	25 July
Millet NDWI	0.35 (BBCH 6)	0.38 (BBCH 7)	0.23 (BBCH 8)
Corn NDWI	0.28 (BBCH 3)	0.35 (BBCH 6)	0.34 (BBCH 7)
Millet NDVI	0.80 (BBCH 6)	0.80 (BBCH 7)	0.65 (BBCH 8)
Corn NDVI	0.78 (BBCH 3)	0.80 (BBCH 6)	0.78 (BBCH 7)
2020	22 June	22 July	11 August
Millet NDWI	0.18 (BBCH 6)	0.37 (BBCH 8)	0.23 (BBCH 9)
Corn NDWI	0.40 (BBCH 3)	0.36 (BBCH 6)	0.30 (BBCH 8)
Millet NDVI	0.62 (BBCH 6)	0.77 (BBCH 8)	0.63 (BBCH 9)
Corn NDVI	0.86 (BBCH 3)	0.75 (BBCH 6)	0.76 (BBCH 8)
2021	12 June	12 July	6 August
Millet NDWI	0.17 (BBCH 3)	0.34 (BBCH 6)	0.26 (BBCH 8)
Corn NDWI	0.06 (BBCH 3)	0.20 (BBCH 6)	0.22 (BBCH 7)
Millet NDVI	0.58 (BBCH 3)	0.77 (BBCH 6)	0.74 (BBCH 8)
Corn NDVI	0.42 (BBCH 3)	0.62 (BBCH 6)	0.68 (BBCH 7)

BBCH 3 = stem elongation; BBCH 6 = flowering; BBCH 7 = milky ripe; BBCH 8 = full ripening; BBCH 9 = senescence.

In 2019, NDVI and NDWI values for millet were extremely close to those of an irrigated corn when comparing data at the same phenological stage; in particular, values at

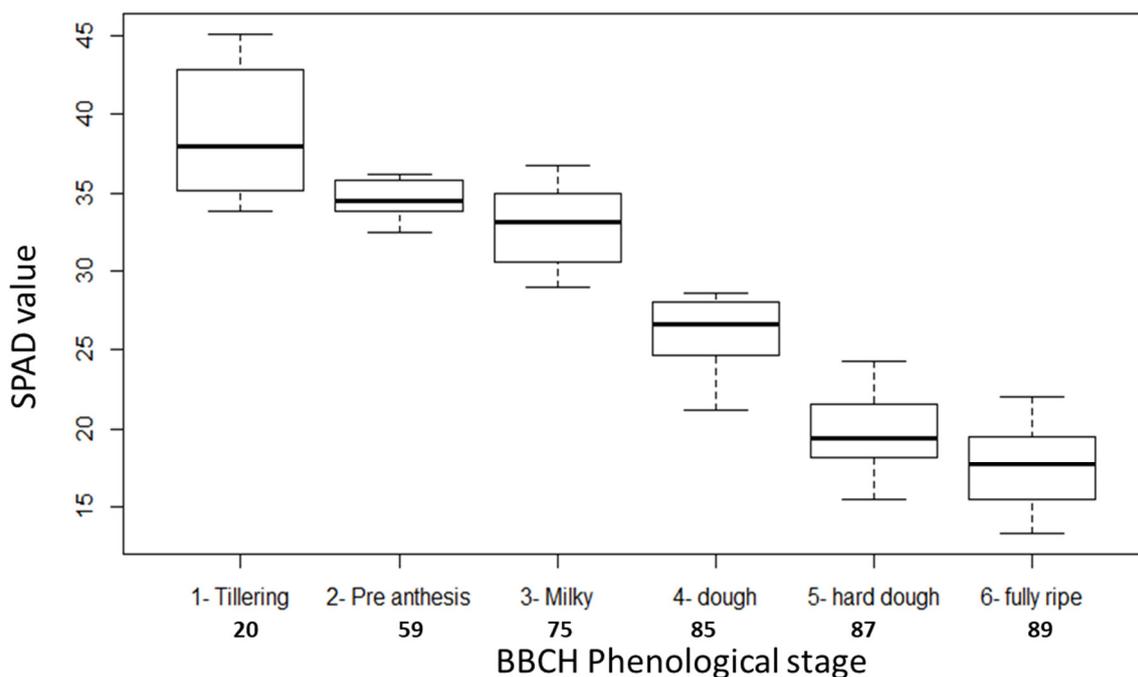
BBCH principal stage 6 (flowering) were perfectly overlapping (0.35 for NDWI and 0.8 for NDVI), but differences remained really restrained during BBCH principal stage 7 (milky ripe) (and a bit in favor of millet, which shows slightly higher values). Millet index values started lowering, as predicted, with the reaching of maturity, when leaves physiologically dry out. In 2020, lower NDVI and NDWI values were recorded for millet at the end of spring compared to the previous year, but the reduction was not so heavy as to show clear water stress. The indexes then settled at high values during summer (once again higher than corn at the same date and at the same BBCH stage), and then fell again at the end of the cycle, in line with what was observed in the previous year. In 2021, millet index values were quite low in June but then greatly improved in summer, surpassing those of irrigated corn. The particularly low values of corn indexes in June are due to the fact that the crop had not yet completely closed the row, and the presence of bare soil significantly reduced the NDVI and NDWI values.



**Figure 4.** NDVI and NDWI indexes calculated on 5 July 2019 for millet (VM) and corn (AC).

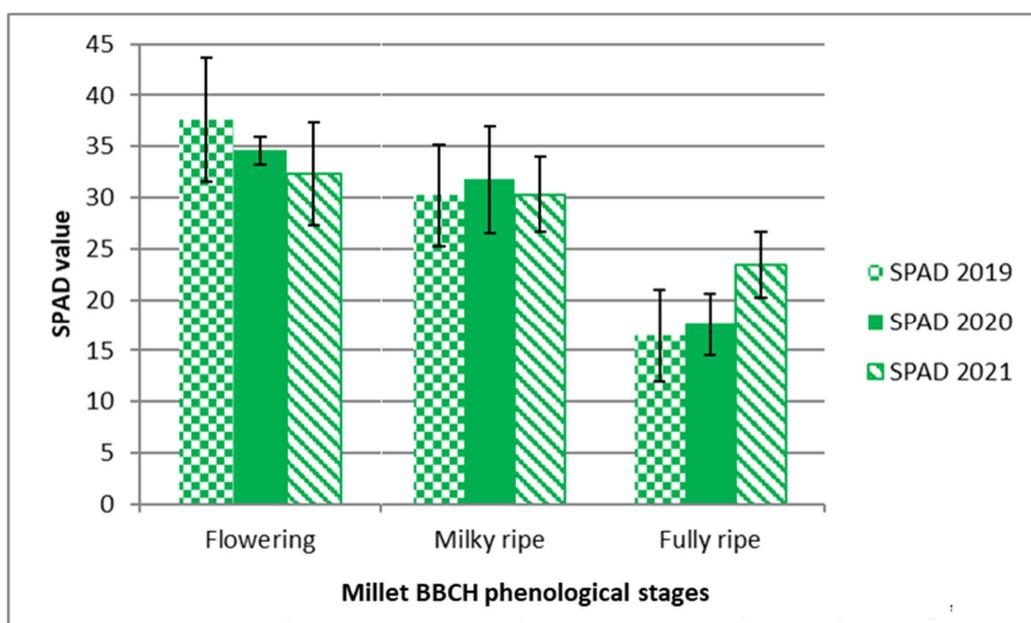
### 3.4. Spad

Figure 5 reports a box-and-whisker plot of SPAD measurements on proso millet during the 2020 life cycle. The “box” is bounded by the first and third quartiles, divided inside by the median. The segments (“whiskers”) are delimited by the minimum and maximum of the values.



**Figure 5.** SPAD measurement on proso millet during the 2020 life cycle at the VM site.

SPAD values drop throughout the life cycle, from 38.9 during BBCH principal stage 2 (tillering, contemporary in millet to stem elongation-BBCH principal stage 3, see Ventura et al., 2020) to 17.6 at BBCH 89 (full maturity), in accordance with crop progressive senescence as the life cycle is completed. Figure 6 shows the good accordance of 2019, 2020, and 2021 SPAD values for the main phenological stages. Therefore, in all experimentation years, millet SPAD values stayed high until flowering, indicating optimal crop vigor, without any clue of drought stress, and then dropped in the last phenological stages, in line with a normal drop in physiological parameters associated with crop senescence [30].



**Figure 6.** SPAD values at the same phenological stages at the VM site in 2019, 2020, and 2021. Bars indicate the standard deviation.

### 3.5. Agronomic Performances

Millet had a good agronomic performance at the VM site open field in the three years of experimentation (Figure 7). The crop homogeneously and abundantly covered the soil, showing excellent competition with weeds. The mean results of the agronomic survey, carried out before harvesting on 3 separate sampling areas of 1 m<sup>2</sup>, are presented in Table 4.

**Table 4.** Agronomic parameters collected at the VM site, expressed as the mean of 3 separate sampling areas of 1 m<sup>2</sup>. For yield/panicle, plant height, and panicle length, formal statistical tests were performed. Different letters indicate significant differences ( $p < 0.05$ ).

Parameter	VM Site (Mean of 3 Separated m <sup>2</sup> )		
	2019	2020	2021
Ground cover (%)	93	83	70
Weeds presence (%)	12	8	23
Yield/panicle (g)	3.1 ± 1.1 a	3.6 ± 1.2 a	3.1 ± 1.1 a
Plant height (cm)	82.2 ± 5.3 a	100.5 ± 6.5 b	84.2 ± 6.4 a
Panicle length (cm)	33 ± 6.5 a	23.6 ± 4 b	17.8 ± 2.5 c

For panicle length transformed via logarithmic transformation, ANOVA highlighted significant differences among the 3 years ( $p < 0.05$ ). Panicle length in 2020 was significantly decreased with respect to 2019, and in 2021, it was significantly shortened with respect to 2020, as highlighted by Tukey's test ( $p < 0.05$ ). Height data were normally distributed, but they were not homoscedastic. Therefore, the non-parametric Welch test was performed, and the Duncan–Waller test was used to separate the means. Plant height at panicle insertion showed significant differences among the three years, as revealed by the Welch test ( $p < 0.05$ ). Specifically, it was significantly greater in 2020 compared to 2019 and 2021. In 2019 and 2021, differences in plant height were not significant. Regarding the yield/panicle, the Kruskal–Wallis test did not show any significant difference ( $p > 0.05$ ), so there was no difference in yield/panicle in the 3 years of experimentation. Therefore, panicle length significantly decreased, passing from 2019 to 2020 and then to 2021, while plant height appeared significantly lower in 2020 compared to the other two years of experimentation. However, these differences did not produce any difference in yield per panicle, which was unchanged over the 3 years.



**Figure 7.** VM site open field proso millet in 2019 (a), 2020 (b), and 2021 (c) during the ripening stage (BBCH 7-8).

### 3.6. Water Balance and WUE

Proso millet and corn grain yield and water balance for 2019, 2020, and 2021 seasons are reported in Table 5. The table also reports the calculation of crop WUE, expressed as the ratio between grain production (kg dry grain/m<sup>2</sup>) and water consumption (m<sup>3</sup>/m<sup>2</sup>), as

the mean of all separated sampling areas. Regarding the period from sowing to BBCH 16 (six leaves fully expanded), calculations confirm that the net water loss from the soil system compared to the water gained from precipitation was zero (2019) or negligible (2020 and 2021). So, the decision to compute millet water balance from BBCH 16 was solid.

**Table 5.** Proso millet and corn water balance and WUE in 2019, 2020, and 2021 (VM and AC site, respectively).

	Proso Millet 2019	Proso Millet 2020	Proso Millet 2021	Corn 2019	Corn 2020	Corn 2021
Grain production (0% UR) (kg m <sup>-2</sup> )	0.34	0.35	0.21	1.44	1.48	1.33
I (mm)	0	0	0	180	190	213
P (mm)	77.0	139.4	69.0	334.2	219.4	126.8
UP (mm)	39.1	34	20.2	126.3	25.4	20.2
Δθ (mm)	48.3	18.6	45.3	-21.1	36.0	60.6
RUN (mm)	0	0	0	0	0	0
PREC (mm)	0	0	0	0	0	0
ETc (mm)	164.4	192	134.5	619.4	470.8	420.6
ETc (m <sup>3</sup> m <sup>-2</sup> )	0.16	0.19	0.13	0.62	0.47	0.42
WUE (grain kg/water m <sup>3</sup> )	2.07	1.83	1.58	2.33	3.15	3.17

#### 4. Discussion

The aim of this study was to assess the contribution offered by proso millet to enrich agricultural sustainability and resilience to climate change in Italy and on the European side of the Mediterranean Basin, to reduce water consumption for cereal production, and to offer a new resource to farmers, for whom corn production becomes less and less reliable. During experimentation, millet faced three difficult seasons from an agronomic point of view, in the full manifestation of the uncertainties caused by climate change. Crop season 2019 was characterized by a quite cold spring, followed by heat waves in June and July. The 2020 season, on the other hand, was characterized by an extremely dry spring. In 2021, drought and heat waves manifested simultaneously in June and July. The extremely different meteorological trend in the period following sowing could have influenced the first stages of development, explaining at least partially the difference in the 107 degrees days necessary to reach BBCH 16 in 2019 and 2020. For the rest of the life cycle, the thermal thresholds identified in 2019 for the achievement of subsequent phenological stages have been largely confirmed, suggesting that these thresholds are consistent.

NDVI and NDWI vegetative indexes calculated on dry-grown millet and on traditional irrigated corn have allowed us to make some interesting considerations. In 2019, although no irrigation was used, millet NDVI was never less than 0.8 until the ripening phase (BBCH principal stage 8) and then settled at 0.65 (in any case a high value, considering natural crop senescence in the final stages of the life cycle). The same consideration can be made, in qualitative terms, for NDWI; index values were 0.35 and 0.38 at the flowering and grain development stages, respectively, showing no water stress for the crop, despite summer heat waves. The index drop to 0.23 appears to be in line with senescence given by the achievement of the final phases of the life cycle. We can therefore state that millet showed high vigor and no signs of water stress during the 2019 growing season. The ability of this crop to effectively resist hot summers without any need for irrigation emerges even better if we compare NDVI and NDWI calculated on millet with those calculated on conventional irrigated maize at the same phenological stages. The indexes of the two crops are in fact widely matching (and slightly in favor of millet during the grain

development stage), despite the fact that corn could take advantage of 180 mm of irrigation during the crop cycle.

In 2020, the extreme drought in spring resulted in lower NDVI and NDWI indexes (although not severely deficient) until BBCH principal stage 6 (flowering) compared to the previous year. However, millet proved to be able to effectively take advantage of summer rainfall, showing July NDVI and NDWI values of 0.77 and 0.37, respectively, perfectly overlapping those of irrigated corn on the same date. The subsequent decline in the indexes at the end of the season appears to be in line with the physiological senescence of the crop and perfectly comparable to the decline observed in the survey of the previous year. Therefore, despite the drought in spring, proso millet was able to recover successfully, taking advantage of summer precipitation and showing good resilience. Maize, in the same season, was supported by 190 mm irrigation, without which it would have presumably shown lower indexes and yield.

In 2021, as in 2020, spring water scarcity resulted in quite low NDVI and NDWI up to anthesis, compared to 2019. Then, the crop greatly improved its vigor and water status, confirming its resilience to adverse meteorological conditions, reaching NDVI and NDWI values of 0.77 and 0.34, respectively, in the month of July, showing higher values than those of irrigated corn.

With regard to SPAD values, we found that chlorophyll content decreases over the life cycle as full ripening is reached. This is in accordance with the normal crop physiology, without indicating that the crop was in a water stress condition. In fact, [30] also observed a progressive reduction in proso millet chlorophyll content starting from the grain filling stage. Moreover, the mean SPAD value we observed in the first part of the life cycle (close to 40), when millet was still green and in full photosynthetic activity, was quite close to SPAD values observed by [31] on proso millet ( $44.4 \pm 0.5$ ) cropped in a 100% ETm (maximum crop EvapoTranspiration) water restitution regime between 34 and 48 DAS (days after sowing). The proximal SPAD measurements confirm what was observed through the vegetative indexes obtained from SENTINEL-2; proso millet, despite three growing seasons characterized by summer heat waves, extremely dry spring, and concomitant drought and heat waves, was able to sustain satisfactory photosynthetic activity, without suffering from water stress. The progressive reduction of SPAD values, which can be easily overlapped in the three years of experimentation, seems to be in accordance with normal crop physiology, without indicating a stressful condition.

The only indication of a possible partial effect of water stress on millet in 2020 and 2021 is given by the significantly shorter panicle length compared to 2019. In fact, as reported by [32], panicle length in cereals is one of the traits that can be affected by water stress. However, the reduction of this trait generally appears contained, and the effect of this reduction on yield is also contained. In fact, in cereals such as millet, when water stress causes a strong reduction in grain productivity, this is mainly due to a drastic drop in terms of grain number per panicle and grain weight, instead of panicle length [13,33]. The effect on yield of panicle length reduction appears to be very limited [32]. This is confirmed by our measurements, as yield per panicle showed no significant difference among 2019, 2020, and 2021. In particular, the 2021 season was characterized by a very low level of precipitation in the period from mid-leaf development (BBCH 13) to the end of flowering (BBCH 69) (only 21.6 mm of total precipitation), combined with 3 heat waves, with temperature peaks over 35 °C in the period from stem elongation (BBCH 32-33) to the end of heading (BBCH 59). Thus, proso millet was subjected in this season to drought and thermal stress, higher than in the previous two years. The effects of these stressors may be found in a significant reduction in panicle length.

Other sources in the literature show a reduction of proso millet WUE in conditions of drought stress [13,14]. However, considering that yield per panicle in 2021 is completely comparable to the two previous years and considering that reduction in panicle length is known to cause a negligible yield loss in millet, it is conceivable that the reduction in WUE observed in 2021, due to a decrease in  $\text{kg m}^{-2}$  dry matter grain production, is not the sign

of a stressed crop but could be mainly caused by a higher weed presence and a lower ground cover rate compared to 2019 and 2020. In fact, there are no evident signs of stress in SPAD values and satellite indexes, which were highly satisfactory during the summer.

Moreover, it is interesting to note that, although it is well known that drought stress causes a reduction in plant height in cereals [34], no significant difference was observed for this trait between 2019 and 2021. On the contrary, plant height appears significantly higher in 2020, despite a notably drier spring than in 2019. It is therefore conceivable that the effect of water stress on plant height in millet is variable, depending on the intensity and the moment in which the stress occurs.

Considering the average proso millet grain yield in Southern Europe (1.8 t/ha-FAO-STAT), we can state that proso millet showed a good performance during this three-year experimentation, despite three differently adverse agricultural years, especially if we consider that not only was the crop never irrigated, but no other productive inputs or cultivation treatments were used. Regarding WUE, this remained satisfactory, as well as the yield, showing values around  $2 \text{ kg m}^{-3}$  in 2019 and 2020 and slightly above  $1.5 \text{ kg m}^{-3}$  in 2021, corresponding to  $20 \text{ kg ha}^{-1} \text{ mm}^{-1}$  and  $15 \text{ kg ha}^{-1} \text{ mm}^{-1}$ , respectively. In the FAO Thematic Report “Status of water use efficiency of main crops” [35], values of WUE up to  $12 \text{ kg ha}^{-1} \text{ mm}^{-1}$  are reported for dry land millet in Sahel (semi-arid African region), and in a long-term experiment in the semi-arid Central Great Plains of the United States (ideally suited for dry land millet production) [36], values ranging from 8.37 to  $33.62 \text{ kg ha}^{-1} \text{ mm}^{-1}$  were recorded. Therefore, the results obtained during the experiment were satisfactory, if compared with the data available in the literature, despite adverse and different meteorological trends.

Regarding maize, it was cropped following the best agronomic techniques available and irrigated with efficient and calibrated water use, thanks to the indications produced by the IRRIFRAME platform. This careful agronomic management allowed us to obtain very satisfactory yields in the three-year experimentation and WUE values in line with what is reported by the FAO Thematic Report. In 2021, summer NDVI and NDWI values were lower than the previous two years, as 2021 was the experimentation year with the lowest rainfall amount.

The higher WUE values in corn were due not only to much greater agronomic inputs on the crop, but to the fact that modern corn hybrids are the result of decades of selection and breeding, unlike millet, whose biodiversity potential offered to researchers and farmers is still to be explored.

## 5. Conclusions

The results of the present study show that proso millet is able to adapt to different climatic conditions, maintaining stable yields and WUE. The three years of experimentation differed in terms of meteorological data: 2019 had excessive rainfall during spring, 2020 was characterized by drought conditions, especially in late winter and spring, and 2021 registered high temperatures in summer. In this variable scenario, the stability of millet’s performance confirmed a high resilience potential toward climate change impacts. This high adaptability to different environmental conditions might motivate a reintroduction of this crop in Northern Italy (and the Mediterranean basin in general) rotations, as an alternative crop of maize, which is the summer crop most affected by climate change in this area, for both agronomic and phytosanitary reasons. Proso millet certainly represents a resource for farmers, considering its short life cycle, which allows it to be employed as a catch crop and to avoid drought conditions in late summer. In addition, proso millet showed vegetational index values and a water status completely comparable to conventional irrigated and fertilized corn. The conclusions of the present study demonstrate the high future potential of proso millet as a resilient and valuable crop for the Mediterranean region from different points of view. The agronomic performances discussed in this research, together with the interesting nutritional properties (mineral content and gluten-

free grain), enhance the potential markets for proso millet, especially for human consumption. Further investigations could improve the current knowledge about proso millet performance, especially realizing a multi-location trial, which will study the agronomic and grain quality performance of proso millet in different pedoclimatic conditions. Finally, future studies are needed in order to explore the genetic diversity of proso millet and to discover the best genotypes for specific environmental conditions.

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