



## Article

# Beyond Beer: Hop Shoot Production and Nutritional Composition under Mediterranean Climatic Conditions

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**Abstract:** For hop growers, surplus shoots are generally a useless by-product of cultivation. Conversely, they may represent a valuable resource due to rising interest towards healthy and traditional foods. A field trial was carried out in Central Italy to characterize shoot production (number of emerged shoots, shoot fresh weight, marketable shoot yield, and shoot diameter) of three commercial hop cultivars (Cascade, Challenger, and Hallertauer Magnum) and to survey shoot proximate composition (ash, ether extract, crude protein, and crude fiber). Green shoots were harvested when they were from 20 to 40 cm in length. The results from two years showed that there was significant difference among the varieties and between growing seasons, both for yield traits and for nutritional composition. H. Magnum showed the highest marketable shoot yield (152 g per plant, two-year mean), while Cascade had the best proximate composition. The number of emerged shoots per plant varied from 62.5 of Cascade to 84.3 of H. Magnum over a two-year average. Marketable shoot yield showed a positive relationship with number of shoots and average shoot fresh weight, while no significant correlation was found with shoot diameter. Hop shoots proved to be a low-fat food (ether extract from 2 to 6% dry matter (DM)) with high protein (from 22 to 30% DM) and fiber content (from 10 to 16% DM).

**Keywords:** hop; *Humulus lupulus* L.; shoot yield; proximate composition; Cascade; Challenger; Hallertauer Magnum

## 1. Introduction

Hop (*Humulus lupulus* L.) is a dioecious perennial climbing plant, and it is known mainly for its use in the brewing industry.

Even though hop can grow between 35 and 50° latitude in both hemispheres, commercial production is traditionally limited to moist and temperate regions like central Europe and the Pacific Northwestern United States [1–3]. In 2018, the world's top hop-producing countries were USA, Germany, and China; however, the USA and all European countries together shared about 88% of world production [4].

While hop today is mainly grown for its female inflorescence (commonly known as cone but formally defined as strobilus), it actually has a long history of being used for various medicinal, household, and culinary purposes [5,6]. Similarly to asparagus, young hop shoots are eaten in spring, which is one of the oldest and traditional uses of the plant [7]. Hop shoots are a delicate vegetable

greatly sought after in most European countries [8]. In the Mediterranean region, foraging for wild hop shoots is quite popular, thus different ways to cook them have been originated [9–11]. After boiling, young shoots have low fat content ( $<0.2$  g/100 g), energy value (25 kcal/100 g), and sodium content ( $<40$  g/100 g), whereas they are a good source of dietary fiber and vitamin B9 [12–14]. Moreover, wild hop shoots have less than 67 mg/100 g of oxalic acid and high vitamin C levels (about 40 mg/100 g), and thus they are a potential source of new functional ingredients in developing new foodstuffs [14]. Fresh green shoots have a short shelf life, so they should be consumed shortly or processed to store them safely for a longer period (e.g., pickled hop shoots).

Developing wild edible plants through cultivation may be a winning strategy to meet the demand of niche markets [15–17] and to limit overharvesting and loss of biodiversity [18–20].

Regardless of this commercial, nutritional, and conservation value, yield potential and nutritional characteristics of wild edible plants are not yet fully explored, particularly in the Mediterranean region [21].

Regarding shoot production in hops, many young shoots emerge from the mature overwintering rootstock in early spring (Figure 1). Bine selection (from two to six per hill) and training are two critical agronomic practices to optimize cone yield [8]. For growers and brewers, surplus shoots are a worthless product of the hopyard, but for gastronomes they are among the world's most expensive vegetables. Their remarkable commercial value is due to limited availability (a few days in spring) and the onerous harvest. Additionally, in a study conducted in Northern Italy, results pointed out that wild hop shoots represent a new source of flavonols and can be incorporated in the diet as a functional food or applied in the nutraceutical ambit [22]. Moreover, in Slovenia, white shoots from cultivated hop were found to be better antioxidants than hop cones and leaves, without any problems of pesticide residues [23]. Finally, use of young shoots as vegetables may represent a valuable additional source of income for hop growers.



**Figure 1.** Hop shoots emerging from a rootstock.

Recently, proliferation of microbreweries in countries that were not typical hop producers (such as those around the Mediterranean basin) has led to (i) an increasing trend in the consumption of special beers and (ii) a growing interest in hop cultivation. In these new growing areas, very limited knowledge is available on cultivated hop, though this species often belongs to local spontaneous flora. Furthermore, while many studies have been conducted to investigate factors affecting cone yield and quality [24–27], information is scarce on shoot production and nutritional composition. According to our best knowledge, few studies aiming to characterize shoot yield under Mediterranean climatic condition were conducted on wild hop [12,17,28] and only one on cultivated hop [7]. This latter study was conducted on young plants (2- and 3-year-old rootstocks), thus making necessary a further investigation to understand rootstock growth over time.

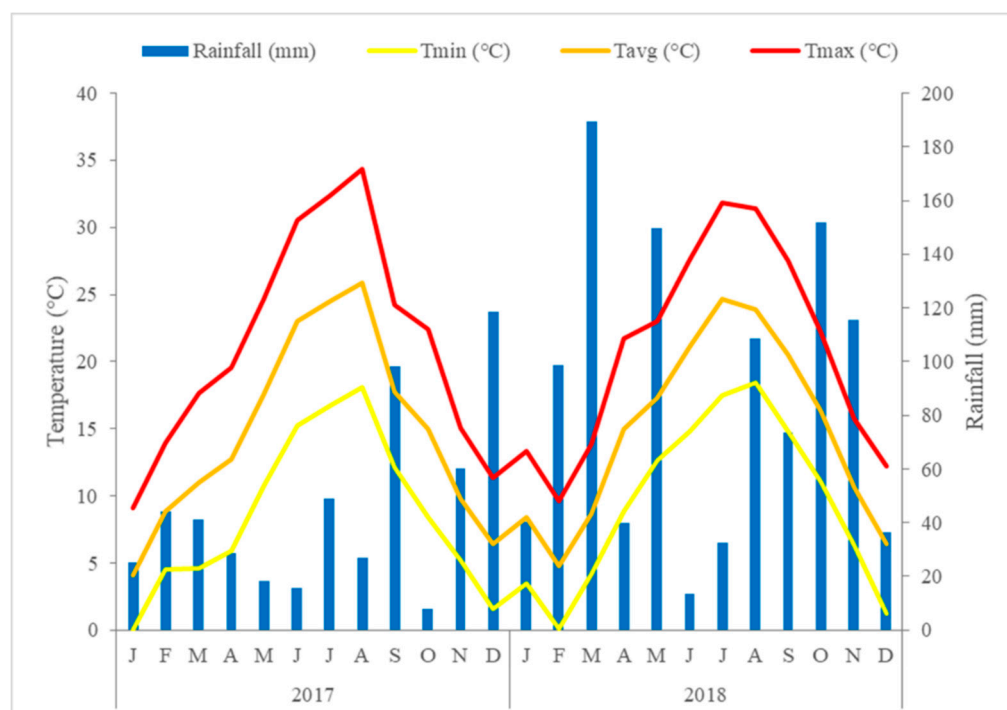
The aim of the present study was to accurately characterize the shoot yield gathered from cultivation of three commercial hop varieties in two subsequent years, under Mediterranean climatic conditions.

Additionally, we surveyed shoot proximate composition to provide novel data for a nutritional evaluation of the crop. Results from this study can help hop growers to better exploit the economic potential of the hopyard, taking into account alternative uses of this versatile crop.

## 2. Materials and Methods

### 2.1. Location, Experimental Design, and Hopyard Management

Field trials were carried out in Viterbo (42°26' N, 12°04' E, altitude 310 m above sea level) in 2017 and 2018 growing seasons. The site has a typical Mediterranean climate, with mean annual air temperature of about 14.5 °C and total annual rainfall of 790 mm. For both years, weather data were retrieved from the meteorological station located within 200 m from the experimental site, and they are reported in Figure 2.



**Figure 2.** Monthly mean maximum (Tmax), mean minimum (Tmin), and average (Tavg) air temperature and rainfall during 2017 and 2018 growing seasons, as retrieved from the weather station of the experimental farm of the University of Tuscia, Viterbo (Italy).

The experimental design was a randomized complete block with three replicates; treatments were varieties. The hopyard was constructed in the spring of 2011 using a standard high trellis system with a finished height of 8 m. Aircraft cable was used for trellis wires. The soil was tilled with a moldboard plow, tilled again with a rotary tiller, and then planted with two hop rhizomes per hill (hereafter referred to as plants) on 13 April 2011. Plants were distanced 1.5 m apart, and rows were spaced at 1.8 m. Each plot consisted of five consecutive plants. All varieties were planted in 2011. A rate of 150 kg ha<sup>-1</sup> of K<sub>2</sub>O was applied before soil tillage and 80 kg ha<sup>-1</sup> of P<sub>2</sub>O<sub>5</sub> at planting time, while N was yearly split in two rates of 50 and 50 kg ha<sup>-1</sup> for spring (late March–early April) and late spring (May–June) applications. The hopyard was not irrigated before shoot collection. Each year, rows were trained with two plastic strings per plant, with two to four of the most vigorous bines trained per string. During each growing season, weeds, pests, and pathogens were chemically controlled.

## 2.2. Plant Materials

Female plants of the three following cultivars were used: Cascade, Challenger, and Hallertauer Magnum (hereafter referred to as H. Magnum). The varieties were selected from a wider list to include a different range of earliness, origin, and brewing quality traits.

Their origin, indicative harvest time, and brewing use are reported in Table 1.

**Table 1.** List of hop cultivars used for the experiment, their harvest time, brewing use, and origin.

Cultivar	Harvest Time	Brewing Use	Origin
Cascade	M	Dual purpose	US
Challenger	L	Dual purpose	UK
Hallertauer Magnum	L	Bittering	Germany

Harvest time: M = medium; L = late.

## 2.3. Field Measurements

Shoots of all cultivars were harvested after completion of bine training. At harvest time, young shoots generally had 5 or 6 nodes completely differentiated, and they were from 20 to 40 cm in length. To avoid border effects, only shoots from the three central rootstocks in each plot were used to assess marketable yield per plant (number of shoots and their fresh weight). Fresh shoots from each rootstock were counted, labeled, cut at the marketable length of 20 cm, and weighed. The average fresh shoot weight was determined by dividing the total fresh weight of shoots by their number. A sub-sample of 10 shoots per replication was randomly chosen for determination of single fresh weight and diameter. Shoot diameter was measured in the median portion of shoots using a digital caliper.

## 2.4. Laboratory Analysis

The proximate composition of hop shoots was determined for the different cultivars. All analytical determinations were performed twice. Gravimetric measurements were carried out by using a PE160 analytical scale (Mettler Toledo S.p.A., Novate Milanese, Italy). The dry matter content (DM) of the samples was determined according to the AOAC method n. 934.01 [24]. Gravimetric determination of ash content (ASH) was obtained after sample incineration by a muffle furnace model VISM 96 (Vismara s.r.l., Trezzano sul Naviglio, Italy) (AOAC method n. 942.05) [24]. Crude protein (CP) was assessed through the Kjeldahl method (AOAC method n. 978.04), and ether extract (EE) was obtained by Soxhlet extraction (AOAC method n. 920.39) [24]. Crude fiber (CF) was estimated through the Weende method (AOAC method n. 978.10) by boiling a 1 g sample aliquot for 45 min in 100 mL of H<sub>2</sub>SO<sub>4</sub> 0.26 N, followed by a forty-five minutes extraction in 100 mL boiling NaOH (0.31 M) in an ANKOM 200 Fiber Analyzer (ANKOM Technology, Macedon, NY, USA) [24]. All compositional data were expressed as percent on dry matter (%DM).

## 2.5. Statistical Analysis

Data were subjected to analysis of variance (ANOVA) using R 3.4.4 software [25] in order to test the main effects of year, cultivar, and their interaction. Significantly different means were separated at the 0.05 probability level by the Fisher's least significant differences test. Simple linear regression analysis was carried out to investigate the relationship between number of shoots and the other variables (e.g., shoot yield, fresh shoot weight, and shoot diameter).

## 3. Results

### 3.1. Shoot Yield Characterization

The number of emerged shoots was not affected by cultivar  $\times$  year interaction, whereas ANOVA showed a significant cultivar ( $p = 0.026$ ) and year ( $p = 7.49 \times 10^{-5}$ ) effect (Tables 2 and 3). In 2017, the number of shoots picked up per plant ranged from 37.3 of Challenger to 74 of Cascade, with an average of 51.7 shoots per plant. In 2018, the average number of shoots was 89.8 per plant, varying from 81.3 of Cascade to 94.7 of H. Magnum. The two-year average showed H. Magnum to be the best performing cultivar, producing 84.3 shoots per plant, while Cascade and Challenger reached a similar result (62.5 and 65.3 shoots per plant, respectively).

**Table 2.** Number of shoots before training, average shoot weight (FW), average shoot weight (DW), marketable shoot yield (FW), and shoot diameter. Year  $\times$  cultivar interaction. Means  $\pm$  standard error.

Year	Cultivar	Emerged Shoots (no. plant <sup>-1</sup> )	Shoot FW (g)	Shoot DW (g)	Marketable Shoot Yield (g plant <sup>-1</sup> )	Shoot Diameter (mm)
2017	Cascade	43.67 $\pm$ 3.38	2.04 $\pm$ 1.14	0.30 $\pm$ 0.018	90.08 $\pm$ 12.31	2.10 $\pm$ 0.07
	Challenger	37.33 $\pm$ 10.87	1.48 $\pm$ 0.06	0.18 $\pm$ 0.009	56.56 $\pm$ 19.04	1.25 $\pm$ 0.03
	H. Magnum	74.00 $\pm$ 7.94	2.77 $\pm$ 0.02	0.29 $\pm$ 0.003	205.21 $\pm$ 23.76	1.91 $\pm$ 0.05
2018	Cascade	81.33 $\pm$ 3.38	1.02 $\pm$ 0.03	0.08 $\pm$ 0.003	82.74 $\pm$ 1.21	1.16 $\pm$ 0.01
	Challenger	93.33 $\pm$ 4.98	0.92 $\pm$ 0.07	0.08 $\pm$ 0.007	86.26 $\pm$ 7.37	1.40 $\pm$ 0.02
	H. Magnum	94.67 $\pm$ 8.21	1.04 $\pm$ 0.07	0.09 $\pm$ 0.007	99.08 $\pm$ 12.90	1.65 $\pm$ 0.07
ANOVA signif.		ns	***	***	***	***
LSD ( $p < 0.05$ )		-	0.25	0.03	0.25	0.16

FW: fresh weight; DM: dry matter; ANOVA signif. codes: 0 '\*\*\*' 0.001; ns: not significant.

**Table 3.** Number of shoots before training, average shoot weight (FW), average shoot weight (DW), marketable shoot yield (FW), and shoot diameter. Cultivar and year mean values. Means  $\pm$  standard error.

Treatments	Emerged Shoots (no. plant <sup>-1</sup> )	Shoot FW (g)	Shoot DW (g)	Marketable Shoot Yield (g plant <sup>-1</sup> )	Shoot Diameter (mm)
Cultivar					
Cascade	62.50 $\pm$ 8.69	1.53 $\pm$ 0.24	0.19 $\pm$ 0.048	86.41 $\pm$ 5.77	1.63 $\pm$ 0.21
Challenger	65.33 $\pm$ 13.62	1.20 $\pm$ 0.13	0.13 $\pm$ 0.024	71.41 $\pm$ 11.29	1.32 $\pm$ 0.04
H. Magnum	84.33 $\pm$ 6.89	1.91 $\pm$ 0.39	0.19 $\pm$ 0.045	152.15 $\pm$ 26.63	1.78 $\pm$ 0.07
ANOVA signif.	*	***	***	***	***
LSD ( $p < 0.05$ )	16.17	0.18	0.02	34.18	0.12
Year					
2017	51.67 $\pm$ 6.93	2.10 $\pm$ 0.19	0.26 $\pm$ 0.019	117.28 $\pm$ 24.42	1.75 $\pm$ 0.13
2018	89.78 $\pm$ 3.62	0.99 $\pm$ 0.04	0.08 $\pm$ 0.004	89.36 $\pm$ 4.97	1.40 $\pm$ 0.07
ANOVA signif.	***	***	***	*	***

FW: fresh weight; DM: dry matter; ANOVA signif. codes: 0 '\*\*\*' 0.001 '\*' 0.05.

As shown in Table 2, a significant cultivar  $\times$  year interaction ( $p = 0.0001$ ) was found for average fresh shoot weight, which ranged from 0.92 g for Challenger in 2018 to 2.77 of H. Magnum in 2017. All cultivars showed a significant decrease in average shoot weight, moving from 2017 to 2018. Specifically, shoot weight was lower by 50% for cultivar (cv) Cascade (2.04 vs. 1.02 g), 38% for Challenger (1.48 vs. 0.92 g), and 62% for H. Magnum (2.77 vs. 1.04 g). Despite this, the two-year



average showed that H. Magnum was the top performing cultivars for this trait (1.91 g, Table 3), followed by Cascade (1.53 g) and Challenger (1.20 g).

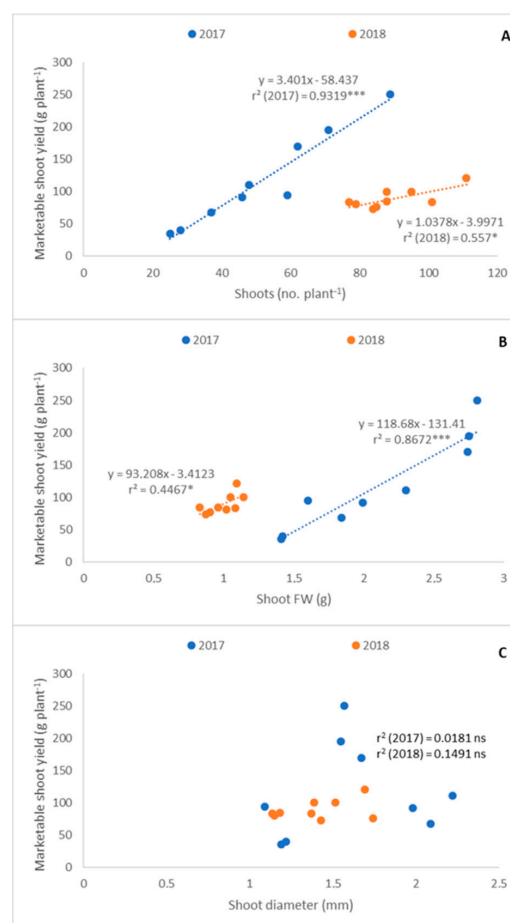
A significant cultivar  $\times$  year interaction was also detected for marketable shoot yield ( $p = 0.0035$ , Table 2).

Shoot yield was not significantly different over the two-year period, except for H. Magnum, whose yield was 52% lower in 2018 than in 2017 (Table 2). The top yielding variety was H. Magnum (about 152 g per plant), while Cascade and Challenger showed similar yields: 86 and 71 g per plant, respectively (Table 3).

Cultivar  $\times$  year interaction significantly affected shoot diameter ( $p = 3.72 \times 10^{-6}$ , Table 2), which ranged from 1.16 to 2.10 mm for Cascade in 2018 and 2017, respectively. Cascade and H. Magnum showed a significant decrease in shoot diameter moving from 2017 to 2018 (−45 and −14%, respectively), while Challenger, in the same period, weakly increased shoot diameter (+12%, not significant).

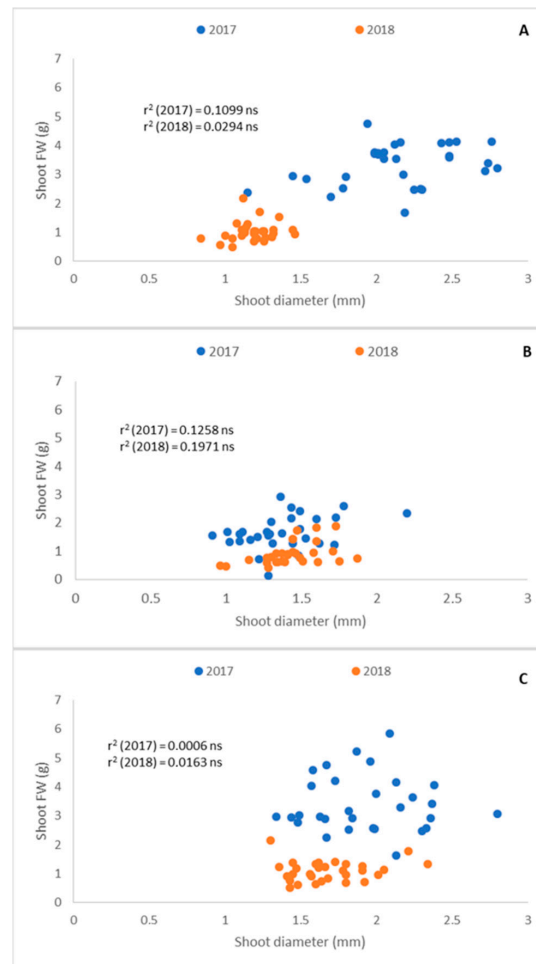
### 3.2. Relationship among Shoot Traits

Figure 3 shows the relationship between marketable shoot yield and number of emerged shoots (A), average fresh shoot weight (B), and shoot diameter (C) in the two growing seasons. Marketable shoot yield was positively and significantly related with both number of shoots per plant and fresh shoot weight in both years, whereas no relation was found with shoot diameter.

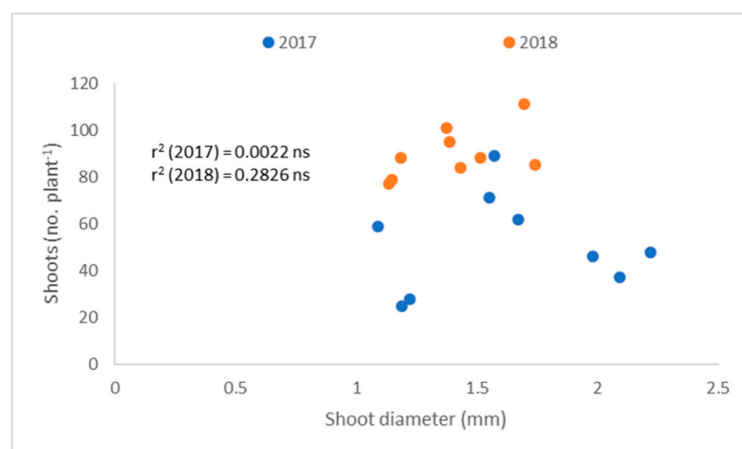


**Figure 3.** Relationship between marketable shoot yield and (A) number of emerged shoots, (B) average shoot fresh weight (FW), and shoot diameter (C) in two growing seasons. Signif. codes: 0 ‘\*\*\*’ 0.001 ‘\*\*’ 0.05; ns: not significant.

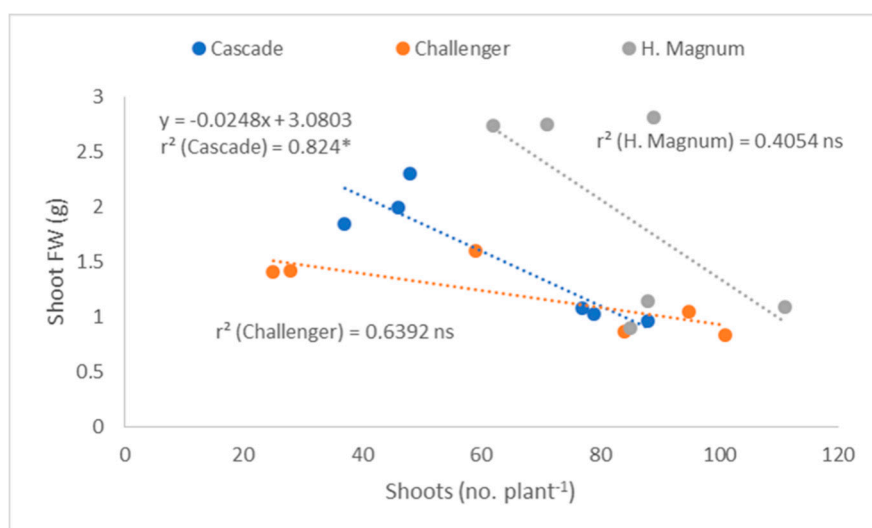
This was because shoot diameter was related neither with number of shoots nor with fresh shoot weight (Figures 4 and 5). Generally, fresh weight of single shoot was negatively related with number of emerged shoots, even though this relation was significant only for Cascade (Figure 6).



**Figure 4.** Relationship between average shoot fresh weight (FW) and shoot diameter of (A) Cascade, (B) Challenger, and (C) H. Magnum in two growing seasons. ns: not significant.



**Figure 5.** Relationship between number of emerged shoots and shoot diameter in two growing seasons. ns: not significant.



**Figure 6.** Relationship between number of emerged shoots and averaged shoot fresh weight (FW) of the three hop cultivars under study. Signif. codes: 0.01 ‘\*’ 0.05; ns: not significant.

### 3.3. Proximate Composition

Proximate analysis and organic acid content of hop shoots are reported in Tables 4 and 5.

**Table 4.** Moisture and proximate composition of hop shoots. Year  $\times$  cultivar interaction. Means  $\pm$  standard error.

Year	Cultivar	Moisture (%)	Ash (% DM)	EE (% DM)	CP (% DM)	CF (% DM)
2017	Cascade	85.5 $\pm$ 0.1	11.64 $\pm$ 0.21	3.88 $\pm$ 0.07	27.04 $\pm$ 0.46	16.48 $\pm$ 0.06
	Challenger	87.6 $\pm$ 0.4	10.56 $\pm$ 0.25	4.03 $\pm$ 0.04	21.56 $\pm$ 0.14	14.15 $\pm$ 0.08
	H. Magnum	89.4 $\pm$ 0.6	10.73 $\pm$ 0.28	6.30 $\pm$ 0.12	26.75 $\pm$ 0.83	12.23 $\pm$ 0.34
2018	Cascade	91.9 $\pm$ 0.05	8.71 $\pm$ 0.18	1.71 $\pm$ 0.02	30.01 $\pm$ 0.14	11.27 $\pm$ 0.28
	Challenger	91.4 $\pm$ 0.05	8.10 $\pm$ 0.14	2.05 $\pm$ 0.02	27.38 $\pm$ 1.50	10.36 $\pm$ 0.15
	H. Magnum	90.9 $\pm$ 0.05	8.17 $\pm$ 0.07	2.61 $\pm$ 0.02	30.00 $\pm$ 0.44	11.26 $\pm$ 0.28
ANOVA signif.		**	ns	***	ns	***
LSD ( $p < 0.05$ )		1.06	-	0.20	-	0.66

Moisture = 100 – DM%; ASH = crude ash; EE = ether extract; CP = crude protein; CF = crude fiber. DM = dry matter; ANOVA signif. codes: 0 ‘\*\*\*’ 0.001 ‘\*\*’ 0.01; ns: not significant.

**Table 5.** Moisture and proximate composition of hop shoots. Cultivar and year mean values. Means  $\pm$  standard error.

Treatments	Moisture (%)	Ash (% DM)	EE (% DM)	CP (% DM)	CF (% DM)
Cultivar					
Cascade	88.68 $\pm$ 1.83	10.18 $\pm$ 0.67	2.79 $\pm$ 0.49	28.52 $\pm$ 0.70	13.88 $\pm$ 1.17
Challenger	89.48 $\pm$ 1.09	9.33 $\pm$ 0.56	3.04 $\pm$ 0.44	24.47 $\pm$ 1.47	12.26 $\pm$ 0.85
H. Magnum	90.13 $\pm$ 0.49	9.45 $\pm$ 0.59	4.45 $\pm$ 0.83	28.37 $\pm$ 0.84	11.75 $\pm$ 0.29
ANOVA signif.	*	***	***	***	***
LSD ( $p < 0.05$ )	0.87	0.37	0.14	1.70	0.46
Year					
2017	87.50 $\pm$ 0.74	10.98 $\pm$ 0.21	4.73 $\pm$ 0.39	25.11 $\pm$ 0.93	14.29 $\pm$ 0.62
2018	91.35 $\pm$ 0.18	8.33 $\pm$ 0.12	2.12 $\pm$ 0.13	29.13 $\pm$ 0.63	10.96 $\pm$ 0.19
ANOVA signif.	***	***	***	***	***

Moisture = 100 – DM%; ASH = crude ash; EE = ether extract; CP = crude protein; CF = crude fiber. DM = dry matter; ANOVA signif. codes: 0 ‘\*\*\*’ 0.001 ‘\*\*’ 0.05.



Generally, shoot moisture was significantly higher in 2018 than 2017. Significant cultivar  $\times$  year interaction was observed. Specifically, shoot moisture was significantly different among the three varieties in 2017 (from 85.5% of Cascade to 89.4% of H. Magnum), while no statistical difference was detected in 2018 (from 90.9% of H. Magnum to 91.9% of Cascade).

Ash content was not affected by cultivar  $\times$  year interaction, whereas ANOVA showed significant cultivar ( $p = 0.00096$ ) and year ( $p = 2.75 \times 10^{-9}$ ) effect. In 2017, ash content of the shoots ranged from 10.6% DM in Challenger to 11.6% DM in Cascade, with an average of 11% DM. In 2018, the average ash content was 8.3% DM, varying from 8.1% DM of Challenger to 8.7% DM of Cascade. The two-year average showed that Cascade shoots had the highest ash content (10.2% DM), while H. Magnum and Challenger showed a similar result (9.5 and 9.3% DM, respectively).

Both EE and CF were affected by cultivar  $\times$  year interaction ( $p = 1.63 \times 10^{-7}$  and  $5.12 \times 10^{-6}$ , respectively), whereas ANOVA showed significant cultivar ( $p = 0.0005$ ) and year ( $p = 7.32 \times 10^{-5}$ ) effect for the CP content. Overall, EE and CF were significantly higher in 2017 than in 2018, whereas CP showed an opposite trend (+16% DM in 2018, averaged over cultivars).

H. Magnum had an EE significantly higher than the other two varieties in both years, followed by Challenger and then Cascade. Crude proteins ranged from 21.6% DM for Challenger in 2017 to 30% DM for H. Magnum and Cascade in 2018. Both the latter cultivars, averaged over years, had a similar CP content (28% DM), significantly higher than that of Challenger (24% DM). Cascade showed the highest crude fiber percentage in 2017, followed by Challenger and H. Magnum, whereas in 2018 H. Magnum and Cascade reached a similar fiber content (11.3% DM), significantly higher than that of Challenger (10.4% DM).

## 4. Discussion

### 4.1. Shoot Production

Comparing results obtained from the present study with those reported by Ruggeri et al. [7] that sampled the same plants but younger (2 and 3 years old), we can obtain interesting information about rootstock growth in the Mediterranean environment. Cascade and H. Magnum increased the average number of emerged shoots by 118% (28.7 vs. 62.5) and 260% (23.3 vs. 84.3), respectively, moving from 2/3-year-old plants (2013 and 2014) to 6/7-year-old plants (2017 and 2018). Challenger was not present in that previous study. In the present study, H. Magnum was the top shoot producing cultivar, whereas sampling younger plants, Ruggeri et al. [7] found Cascade to be the best performing variety. The earlier sprouting, flowering, and maturing cultivar Cascade adapted to the Mediterranean climate faster and better than the other two cultivars [26], and this may have caused an advantage for the growth of rootstock in the early years.

As for average fresh weight of the single shoot, Cascade and H. Magnum increased their average values by 17% (1.31 vs. 1.53 g) and 91% (1.00 vs. 1.91 g), respectively, moving from 2/3-year-old plants (2013 and 2014) to 6/7-year-old plants (2017 and 2018). Moreover, we observed a marked reduction of fresh shoot weight (−53%) moving from 2017 to 2018, while, in the same period, the number of shoots per plant increased by 74%. In the previous study [7], we did not find this negative relationship because the number of emerged shoots varied very little (7%, from 20.5 to 19), and average fresh weight of the single shoot remained substantially unchanged (from 1.15 to 1.16 g). Our results are in agreement with data recorded in other field experiments on *Asparagus officinalis* L. carried out in southern Italy [27]. Conversely, average spear weight of wild asparagus (*Asparagus acutifolius* L.) was found not to be affected by spear number [15]. We compare our results with those obtained on asparagus because no characterization of shoot production was found in the literature for cultivated hops. In our study, the mean weight of 20 cm shoots averaged over years (from 1.20 g of Challenger to 1.91 g of H. Magnum) was in the range of that found by Molina [28] in Spain, collecting 15–30 cm shoots from wild plants in three years and two sites (from 1.22 to 2.48 g). We expected a higher fresh weight from cultivated hop; however, since the method of collection was very different, we can gather

a simple indication from this comparison. In the study by Molina [28], wild shoots were collected in different days from late March to May, thus causing less competition for rootstock reserves among shoots and probably increasing the average single shoot weight.

The marketable yields gathered in our study (from 71 to 152 g per plant, 2-year mean) were markedly higher than those recorded for white hop shoots harvested in Slovenia [23]. In more detail, H. Magnum yielded 5.9 g DM per plant in Slovenia (3-year mean), while it produced 15 g DM per plant (2-year mean) in the present study (152.15 g fresh weight (FW) per plant at 90.13% moisture). This can be justified considering that white hop shoots are collected before they emerge from the soil, thus being still fragile, shorter, and lighter than green hop shoots. In their study, [23] reported that each plant root system had from 15 to 40 white shoots, with the mean fresh mass of each shoot approximately 1 g. Looking at the variation over a longer time of the marketable shoot yield (from 2013–2014 to 2017–2018), Cascade and H. Magnum increased production by 133% (37.14 g per plant vs. 86.41 g per plant) and 564% (23.3 g per plant vs. 84.3 g per plant), respectively. This enormous variation proves the excellent adaptation of these two cultivars to the Mediterranean climate. As expected, and in agreement with the previous study on younger plants, marketable yield was positively and significantly related to number of emerged shoots [7]. In the present study, we also found a significant and positive relation with the fresh weight of the single shoot that was not detected four years earlier [7]. Unexpectedly, we did not find any relation between the fresh weight of the single shoot and its diameter. The reason has to be searched in hop morphology because this species has bines with a hollow core between nodes. This peculiarity makes the positive relationship normally existing between spear diameter and fresh weight of *A. officinalis* [29] not applicable to hop. In fact, the hop shoots with a higher diameter are often those with larger hollow core, while thinner ones are generally less hollow.

#### 4.2. Proximate Composition

Moisture and proximate composition found in the present study is perfectly in the range shown for wild hop shoots harvested in Spain [30], with the exception of fiber. Tardío et al. [30] reported a range of dietary fiber from 4.35 to 6.42% FW (from 30.5% to 44.9% DM) while, in our study, crude fiber content, averaged over years, varied from 11.8% DM of H. Magnum to 13.9% DM of Cascade. It must be noted that crude fiber is only one component of dietary fiber, primarily composed of cellulose and lignin. In more detail, constituents of dietary fiber include cellulose, hemicelluloses, lignin, gums, mucilage, oligosaccharides, pectin, and other associated minor substances [31]. For this reason, crude fiber may grossly underestimate the actual dietary fiber content in foodstuff, but it can help in making intra-study comparison, as in our case.

Shoot moisture was significantly higher in 2018 than 2017 (91.4 vs. 87.5%). This was probably due to the different groundwater availability before and during shoot emergence. Rainfall amount from February to April 2017 was 114 mm (−37% with respect to the long-term average), while in the same period of 2018 it was more than double (328 mm). For the same reason, shoots picked in 2017 were more fibrous than those harvested in 2018. Reduced water availability was already found to significantly enhance fiber content in vegetables [32,33]. EE was also significantly higher in drought conditions of 2017 than the 2018 growing season. In this regard, it has to be said that various environmental stresses may release a lipid-mediated signal. To mitigate such stresses, enhanced syntheses of lipids are often observed in different plants [34].

Conversely, crude protein content increased in the 2018 rainy season as compared to the 2017 drought season. Previous studies have demonstrated that drought can decrease the protein concentration of plant tissues, mainly for a decrease in nitrogen uptake from the soil [35,36]. The ash content significantly decreased in the 2018 rainy season as compared with the 2017 drought season. We actually expected an opposite trend, as a result of reduced nutrient availability and uptake in the drought season [37]. However, heavy rainfall occurred before and during shoot emergence (+82% with respect to the long-term average), which may have caused a significant soil nutrient leaching, only partially replaced by nitrogen fertilization.

A significant year effect on proximate composition of wild hop shoots was also reported by García Herrera [12] in Spain, but the author did not describe the weather pattern of the two years.

## 5. Conclusions

In the present study, all traits investigated (number of emerged shoots, shoot fresh and dry weight, marketable shoot yield, shoot diameter, moisture, ash, ether extract, crude protein, and crude fiber content) were significantly affected by genotype and year.

Marketable shoot yield was positively correlated with shoot number and fresh weight, whereas no correlation was found with shoot diameter. Averaged over the two years, H. Magnum was the top performing cultivar for all traits analyzed of shoot production, while Cascade and Challenger showed similar results.

As for nutritional composition, hop shoots are confirmed to have low lipid content and to be a good source of proteins and fiber. From this point of view, Cascade seems to be the cultivar with the better-quality traits.

These results, coupled with those on shoot and cone yield reported in previous studies [7,26], suggest that Cascade and H. Magnum adapt well to the Mediterranean climatic condition and could represent the best choice for new hopyard establishment in those environments.

Further studies should deeper investigate the chemical composition of hop shoots deriving from different commercial varieties (organic acid content, flavonols, vitamins, and antioxidant properties) in order to better understand and assess their potential benefits for human health.

Cone production and quality in different growing environments should be also characterized.

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