



# Article Grain Yield and Quality Traits of Durum Wheat (Triticum durum Desf.) Treated with Seaweed- and Humic Acid-Based Biostimulants

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Abstract: The ongoing climate change with increasingly frequent, prolonged drought during the vegetation period is a significant factor affecting production of field crops, including durum wheat (Triticum durum Desf.). One of the approaches to effectively protect plants from drought stress is the foliar application of bioactive substances and selection of appropriate genetic material for specific location conditions. In this study, the impacts of brown seaweed based and humic substance-based biostimulants were researched. The positive impact of bioactive substances on grain yield has been reported in many studies. However, the impact on quality components is questionable and not well investigated. In this study, a highly significant ( $\alpha < 0.01$ ) positive impact of bioactive substances on grain yield was confirmed. The highest grain yield was observed on the fertilized variant with humic substances (4.03 t ha<sup>-1</sup>). When compared to control, there was a high statistically significant difference. The biofertilization impact on quality components was weakly positive in most cases, although without statistical significance ( $\alpha > 0.05$ ). The study included evaluating the interactions biofertilization-weather conditions (BW) and biofertilization-variety (BV). According to the ANOVA results, a highly significant impact in BW on grain yield was found, and in BV, a highly significant impact on protein content, falling number, and gluten content ( $\alpha < 0.01$ ) and significant impact on grain yield and vitreousness were found ( $\alpha < 0.05$ ). Correlation analysis among the monitored parameters was performed. The results that we obtained from the multi-annual field research may contribute to sustainable arable farming in areas with a lack of rainfall during vegetation. By foliar application of bioactive substances, we achieved a significant increase in the yield of durum wheat while maintaining or increasing the quality parameters of the grain.

Keywords: durum wheat; bioactive substances; variety; weather conditions

## 1. Introduction

Durum wheat is the second most important wheat species after common bread wheat, and it is grown worldwide [1]. According to statistics [2], world wheat production is about 765 million tonnes. Common wheat accounts for nearly 95% of wheat production, while durum wheat accounts for the remaining 5% [3]. Durum wheat is characterized by yield instability caused by adverse weather conditions, primarily by irregular water distribution and high temperatures during the grain filling stage [4].

Many different food products can be made from durum wheat grains, including pasta, bulgur, bread, etc. [5]. Semolina and pasta quality depend on harmonic protein concentration ensured by high doses of N fertilization. This requirement is often in conflict with the environment, which has led to the need for better crop nutrition management [6]. Similarly, one of the most dominant agricultural productivity determinants is climate [7],



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**Copyright:** © 2021 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). and drought, as the most significant environmental stress factor, causes a reduction in wheat production [8]. Current predictions indicate temperature increases that will affect food security due to decreasing crop yields [9]. If global warming continues at the current rate, it is assumed that, between 2030 and 2052, the temperature will increase by  $1.5 \,^{\circ}C$  [10]. Currently, agriculture in arid areas is threatened by risks from dry and hot weather conditions, which cause wheat yield reduction [11].

Agricultural systems have progressively focused on organic, sustainable, and ecologic crop production [12]. One of these approaches is plant biostimulants, which can be defined as products that stimulate the plant nutrition process. The aim of their application is to improve the functions of the plant, e.g., nutrient efficiency, abiotic stress tolerance, humification, and much more [13]. The foliar application of biostimulants is widespread in various crop species, e.g., maize [14], sunflower [15], or strawberry [16]. The biostimulants causes the reduction of fertilizers while increasing plant tolerance to biotic and abiotic stresses [12]. Biostimulants can be of various origins, derived from various organic materials and combinations. Available biostimulants contain humic substances, seaweed extracts, amino acids, peptides, inorganic salts, etc. [17]. Kauffman et al. [18] first summarized biostimulants by introducing their classification into three groups based on their source and content: 1) biostimulants with humic substances, 2) biostimulants with hormone-containing products, and 3) biostimulants with amino acid-containing products. One of the ways to relieve stress and support plant growth is the application of biofertilizers made from brown seaweed [19]. Fertilization with fresh seaweed in agriculture is an ancient practice. However, biostimulant effects have been recorded only recently [20]. The use of seaweed in agriculture has many advantages. Applying this substance to crops causes better rooting and higher yields, increases drought and salt tolerance, and enhances photosynthetic activity and resistance to various types of diseases [21]. Moreover, many studies confirmed that humic substances enhance plant growth and physiological processes [22–24]. Due to the activation of the wheat control mechanism through root exudates and the increase in the biodiversity of soil microorganisms, it is possible to achieve a higher yield by applying humic substances [25].

The quality of durum wheat can be evaluated in various ways, and each of them is influenced by production parameters. The stability and potentiality of grain yield are influenced by agronomical quality. Semolina yield, humidity, ash content, and grain impurity are influenced by milling quality. Meanwhile, protein content and gluten quantity and quality are influenced by technological processing [26]. There are varieties on the market known to achieve stable average yields with a wide adaptive capacity to the environment. Moreover, varieties with a high yield potential under specific growing conditions are also bred. These are known as varieties with specific adaptability, and they achieve poor yields under non-target conditions [27]. Several authors have confirmed the specific adaptability of modern cultivars in selected productive growing conditions [28–31].

The condition for profitable cultivation is the right selection of genotype for a specific area. Based on experience and recommendations, three varieties were selected for the experiment, in which quantitative and qualitative parameters were monitored. Especially in dry and warm growing areas, the application of bioactive substances is an important intensifying factor. One of the most widely used substances in the world is extracts from brown seaweed, which have been compared with bioactive substances of humic origin and a control, unfertilized variant.

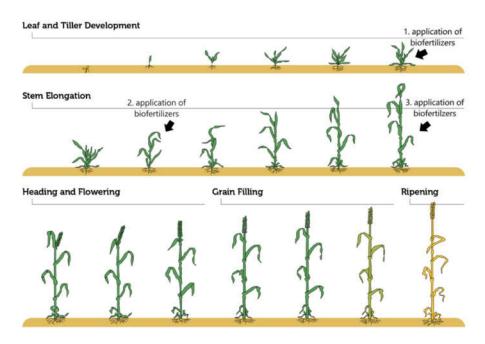
Therefore, in this study, differences in yield and quality of durum wheat caused by foliar application of bioactive substances were researched. All practices and treatments were realized in field conditions, which increases the value, credibility, and practical relevance of the results obtained.

#### 2. Materials and Methods

## 2.1. Locality Description and Climate Characteristics

The experiment was realized under field conditions at a grange in Horný Bar (N 47.947268; E 17.454775). The location is situated on the left bank of the Danube river on Žitný ostrov,

which is a part of the Danubian Lowland. The geological subsoil consists of sand-gravel alluvium. The type of soil is medium-heavy floodplain with low humidity. Weather conditions at the site are typical for the maize crop region, with an average air temperature during the year of +9.3 °C. During the research years, there was no snowfall, and overall, the region is one of the driest areas in Slovakia. According to the long-term climatic normal of the site, the overall precipitation during the wheat vegetation season is about 450 mm (Figure 1).



**Figure 1.** Phenological phases of wheat development. Black arrows indicate the developmental stage of the plant in which biofertilizers have been applied. Figure was adopted and then modified from eKonomics (https://nutrien-ekonomics.com/; 7.3. 2021).

Meteorological variabilities of research years were obtained from Slovak Hydrometeorological Institute, which provided information on daily temperatures and precipitation.

#### 2.2. Plant Material

In the experiment, three varieties of winter durum wheat, namely Auradur (Probstdorfer Saatzucht GesmbH & Co KG, Wien, Austria), Elsadur, and Lunadur (Saatbau Slovensko, s.r.o., Trnava, Slovakia), were planted. Elsadur and Auradur are typical early varieties with resistance to lodging and good qualitative properties. Lunadur has a wide range of planting dates due to its plasticity, has excellent frost resistance, and achieves stable yield. Frost resistance of winter crop varieties is necessary because temperature extremes (cold, frost, etc.) can substantially influence grain yield [32].

## 2.3. Foliar Fertilizers Based on Bioactive Compounds Characterisation

Two treatments with foliar fertilizer application with different bioactive compounds and a control variant were established in the experiment. Biofertilizers Alga 300++P and Alga 300++K based on brown seaweed extract and AminoTotal with high L-amino acids (labeled together as V1) were provided by Agrobiosfer s.r.o. (Bratislava, Slovakia). The company Agrotrade Group spol. s.r.o. (Rožňava, Slovakia) supplied biofertilizers with increased content of humic substances AT-Energia Humín, AT-Mikro Humín, and AT-Úroda with high nitrogen, sulfur, and magnesium contents (labeled together as V2). The biofertilizer treatments were completely compared to control variant (labeled as V0).

## 2.4. Preparation of Field Experiment and Treatments

The forecrop for durum wheat in the experimental years was winter rape (*Brassica napus* conv. *napus*). After the harvest of winter rape, post-harvest stubble plowing by Väderstad Carrier XL (Väderstad AB, Väderstad, Sweden) was conducted. Before the sowing date, the soil samples were taken randomly four times across the experimental site, mixed to the uniform sample, and then analyzed for nutrient content (Table 1). According to the results, fertilizer containing 10% ammonium nitrogen, 26% P<sub>2</sub>O<sub>5</sub>, and 26% K<sub>2</sub>O (Agrofert a.s., Praha, Czech Republic) was applied. The content of N<sub>an</sub> was determined using the colorimetric method, ammonium nitrogen using Nessler's reagent [33], and nitrate nitrogen using phenol 2.4-disulfonic acid [34]. The contents of other macronutrients (P, K, Mg) were determined using the Mehlich III method [35]. The overall size of the experiment was over 21 hectares. One experimental block was established on 7776 m<sup>2</sup> (54 × 144 m), and each variant had three replications. The experiment was established using the method of randomly arranged experimental members.

Table 1. Nutrients content in soil after analysis in autumn.

Veer/Cell Denth		Nutrient Con	tent (mg kg <sup>-1</sup> )	
Year/Soil Depth	N <sub>an</sub>	Р	K	Mg
2016/0.3 m	17.66	56.31	122.70	270.05
2017/0.3 m	22.10	45.28	119.98	290.23

Pre-sowing soil preparation was realized by deep loosening and subsequent alignment of the soil with Farmet Duolent 3m (Farmet a.s., Česká Skalice, Czech Republic). NPK fertilizer was applied before durum wheat planting by Rauch Axis M (Rauch Landmaschinenfabrik GMBH, Sinzheim, Germany) in a dose of 200 kg ha<sup>-1</sup> based on nutrient analysis. Wheat varieties were sown on 6 October 2016 and 4 October 2017 by Väderstad Rapid 4m (Väderstad AB, Väderstad, Sweden). The sowing rate was 4.5 million germinating grains per hectare with 0.125 m inter-row spacing. Herbicides, fungicides, and nitrogen treatments during vegetation were applied as needed and according to the durum wheat cultivation system.

During the vegetation period (Figure 1), fertilizers with bioactive compounds were applied three times by Tecnoma Galaxy 3000 (Tecnoma, Épernay, France). The control variant was treated only with water. It was necessary to spray plants early in the morning because of windless and suitable temperature conditions as in Saa et al. [36]. The concentration and chemical structure of applied molecules, stomatal opening, plant morphology, and several other factors impact the penetration of the compounds into leaves [37].

#### 2.5. Harvesting and Laboratory Analysis of Samples

Each experimental plot was harvested separately by Claas Lexion 780 (CLAAS UK Ltd., Saxham, England) on 6 July 2017 and 3 June 2018, and the value of grain yield (GY) was determined immediately by weighing on a mobile scale. Samples for laboratory analysis of quality were taken from the variants.

The grain moisture (GM) and bulk density (BD) were analyzed by GAC 2100 AGRI (DICKEY-john, Auburn, USA) as in the work of Armstrong et al. [38]. The falling number (FN) was determined using the instrument ED 3000 (EVOL consulting s.r.o., Vydrany, Slovakia) following the standard STN ISO 3093. The vitreousness (V) was evaluated by DURUM TESTER (EVOL consulting s.r.o., Vydrany, Slovakia) on a sample of 200 grains according to the standard STN 46 1011-11. The protein content (PC) and gluten content (GC) respectively were determined using the Perten Inframatic 9500 (Perten Instruments GmbH, Hamburg, Germany), similarly to Büyük et al. [39].

## 2.6. Statistical Analysis Methods

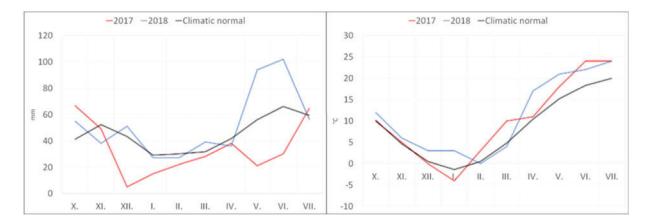
Statistical evaluation of the obtained results from experimental variants, years, and varieties was determined using software Statistica 10 (StatSoft, Inc., Tulsa, OK, USA). First, the three-way ANOVA test evaluated the data (analysis of variance) to determine the significance of the impact of factors (weather conditions, variety, biofertilization) and interactions among them. At significance levels of  $\alpha = 0.05$  or  $\alpha = 0.01$ , the normality of obtained data was examined by Tukey's HSD (honestly significant difference) test. Relationships among all measured traits were determined by Pearson correlation analysis.

## 3. Results

## 3.1. Weather Conditions in Experimental Years

The weather conditions in 2017 and 2018 were very different (Figure 2). When compared to standard climatologic normal (1951–2000), extraordinary below normal precipitation was recorded in December 2017 (-38 mm), while precipitation levels in May (-35 mm) and June 2017 (-36 mm) were very below normal. In contrast, according to Kožnar and Klabzuba [40], precipitation below the normal level was not recorded in 2018. It can be assumed that this was the primary cause of the significant differences in the obtained values of production parameters.

In terms of the sum of temperatures, extraordinary above normal values were recorded, especially in the summer months of both years. The natural development of the durum wheat was endangered in 2017 by the alternation of a below-normal month (January) with above-normal (February) and extraordinarily above-normal (March) months.



**Figure 2.** Precipitation (mm) and temperature (°C) variabilities of research years on experimental area. Included are long-term climatic normal values (1951–2000) of region.

## 3.2. Impact of the Foliar Bioactive Substances Treatment on Grain Yield and Quality Traits

From quantitative traits of durum wheat, grain yield was measured as the primary marker of wheat production. The statistical evaluation showed a highly significant impact of biofertilization only on grain yield (Table 2). The highest mean value of grain yield of the two years was reached on the V1 variant,  $4.03 \pm 1.62$  t ha<sup>-1</sup> (Table 3). Highly significant differences between control, V1 (+0.20 t ha<sup>-1</sup>), and V2 (0.23 t ha<sup>-1</sup>), respectively, were found.

	Production Components					
Source of Variation	GY	PC	V	FN	BD	GC
	p-Values					
W	0.0000 **	0.0000 **	0.0000 **	0.0000 **	0.0360 *	0.0000 **
V	0.0000 **	0.0061 **	0.0000 **	0.0001 **	0.0000 **	0.0000 **
В	0.0000 **	0.2505	0.9131	0.8173	0.6405	0.8771
ВxW	0.0000 **	0.2923	0.8637	0.1280	0.6880	0.9020
B x V	0.0166 *	0.0000 **	0.0355 *	0.0006 **	0.0641	0.0021 **
B x W x V	0.0000 **	0.0002 **	0.8380	0.3285	0.0014 **	0.0000 **

**Table 2.** Analysis of variance (ANOVA) for impact of different source of variation on production components of durum wheat.

W—weather conditions; V—variety; B—biofertilization; GY—grain yield; PC—protein content; V—vitreousness; FN—falling number; BD—bulk density; GC—gluten content. \* and \*\*: significance at  $\alpha \le 0.05$  and at  $\alpha \le 0.01$ , respectively.

The qualitative parameters of wheat grain were not significantly affected by the application of bioactive compounds (Tables 2 and 3). The bulk density, protein content, vitreousness, falling number, and gluten content means of each treatment variant were very similar, without statistically significant differences (Table 3). However, a positive enhancement effect was recorded on biofertilizer variants. The highest values of protein content, vitreousness, and gluten content were achieved on variant V2. In contrast, values of bulk density and falling number were the highest on variant V1

**Table 3.** Yield components of durum wheat in control and in the biostimulant treated variants. Lower case letters (a, b) indicate the significant differences among the variants determined by Tukey's HSD test, included is also standard deviation ( $\pm$ SD). \*\*  $\alpha = 0.01$  indicate level of significance.

	<b>V</b> 0	V1	V2
Grain yield (GY) (t ha <sup><math>-1</math></sup> ) **	$3.80\pm1.83~\mathrm{b}$	$4.00\pm1.82$ a	$4.03\pm1.62$ a
Protein content (PC) (%) **	$15.64\pm1.20~\mathrm{a}$	$15.78\pm1.14~\mathrm{a}$	$15.86\pm1.50$ a
Vitreousness (V) (%) **	$87.22\pm4.52~\mathrm{a}$	$87.17\pm4.25~\mathrm{a}$	$87.39\pm5.25\mathrm{a}$
Falling number (FN) (s) **	$346.56 \pm 63.37$ a	$347.17 \pm 58.02$ a	$343.06 \pm 56.86$ a
Bulk density (BD) (g $l^{-1}$ ) **	$784.06 \pm 33.50$ a	$789.22 \pm 15.78$ a	$785.44 \pm 28.88$ a
Gluten content (GC) (%) **	$34.51\pm2.91~\mathrm{a}$	$34.47\pm3.09~a$	$34.67\pm3.99~\mathrm{a}$

3.3. Interactions between Experimental Factors

3.3.1. Biofertilization x Weather Conditions (BW)

In this experiment, significant weather condition differences between researched years were observed (Figure 1). This was expected, but the objective was to determine the impact of biofertilizers in years with different weather conditions on production components. The BW interaction was significant for all parameters except for the bulk density of grain, although at different levels. A significant impact of BW was found only on grain yield ( $\alpha = 0.01$ ). However, differences between results of other components were visible. As Table 4 highlights, the highest grain yield was identified in the V1–2018 interaction, which was primarily due to the increased total precipitation in the given year. Yield enhancement due to bioactive substances was observed primarily in 2017 in drought stress conditions. The interaction V2–2017 provided a significantly higher grain yield ( $\alpha = 0.01$ ) compared with the non-fertilized variant in the same year. Similar trends were observed on the variant treated with preparations based on brown seaweed. These results are very important because the positive effect of biofertilizers in the area affected by drought stress was confirmed.

## 3.3.2. Biofertilization x Variety (BV)

In the context of significantly different results of varieties in Table 5, it was expected that the resulting values would also be very different in the interaction with biofertilizers (BV). In this study, the high variability of grain yield per hectare (GY) in the BV interaction

case was confirmed. The significantly highest GY (Table 4) was observed in BV interaction V2–Lunadur ( $\alpha = 0.01$ ). This interaction also had the lowest gluten content (GC) and one of the lowest concentrations of protein content (PC). An interesting course of the curves was registered for the falling numbers (FN) of the grains. In all BV interactions, different tendencies were recorded, but based on statistical evaluation, the highest FN was found in the V1–Lunadur interaction. In contrast, when comparing all BV interactions, minimal differences in grain vitreousness values were found, although a highly significant differences were found in BV interactions in terms of bulk density (BD). The BD means of variety Lunadur on all treatment variants was higher than other varieties, however without statistical significance between variants ( $\alpha = 0.05$ ). As Table 4 also highlights, the differences between the varieties were significant, but no statistically significant effect was recorded in combination with biofertilizers.

**Table 4.** Two-way interactions between weather conditions of experimental years, varieties and biofertilization treatments for grain yield (GY), protein content (PC), vitreousness (V), falling number (FN); bulk density (BD) and gluten content (GC). Treatment variants are labeled as V0 (control), V1 (seaweed based) and V2 (humic substances based). Lowercase letters indicate differences according to HSD Tukey test at level of significance  $\alpha = 0.05$  for BD in B x W interaction and  $\alpha = 0.01$  for all others, respectively.

Interac B x V		GY (t $ha^{-1}$ )	PC (%)	V (%)	FN (s)	BD (g l <sup>-1</sup> )	GC (%)
	V0	2.10 <sup>b</sup>	16.70 <sup>b</sup>	91.00 <sup>b</sup>	401.33 <sup>b</sup>	779.11 <sup>a</sup>	37.14 <sup>b</sup>
2017	V1	2.29 <sup>c</sup>	16.66 <sup>b</sup>	90.67 <sup>b</sup>	388.44 <sup>b</sup>	786.56 <sup>a</sup>	37.03 <sup>b</sup>
	V2	2.55 <sup>d</sup>	16.74 <sup>b</sup>	91.11 <sup>b</sup>	396.00 <sup>b</sup>	777.89 <sup>a</sup>	37.12 <sup>b</sup>
	V0	5.51 <sup>a</sup>	14.58 <sup>a</sup>	83.44 <sup>a</sup>	291.78 <sup>a</sup>	789.00 <sup>a</sup>	31.87 <sup>a</sup>
2018	V1	5.70 <sup>e</sup>	14.91 <sup>a</sup>	83.67 <sup>a</sup>	305.89 <sup>a</sup>	791.89 <sup>a</sup>	31.91 <sup>a</sup>
	V2	5.50 <sup>a</sup>	14.98 <sup>a</sup>	83.67 <sup>a</sup>	290.11 <sup>a</sup>	793.00 <sup>a</sup>	32.21 <sup>a</sup>
B x	V	GY (t $ha^{-1}$ )	PC (%)	V (%)	FN (s)	BD (g l <sup>-1</sup> )	GC (%)
	V0	3.62 <sup>b</sup>	15.80 <sup>ab</sup>	84.83 <sup>ab</sup>	344.67 <sup>ab</sup>	752.50 <sup>b</sup>	34.49 bcc
Elsadur	V1	3.87 <sup>c</sup>	15.35 <sup>a</sup>	85.17 <sup>ab</sup>	310.50 <sup>a</sup>	776.50 <sup>abc</sup>	33.03 <sup>ab</sup>
	V2	3.82 <sup>c</sup>	15.48 <sup>a</sup>	84.00 <sup>b</sup>	333.00 <sup>a</sup>	770.50 <sup>ab</sup>	33.30 abo
	V0	4.45 <sup>e</sup>	15.55 <sup>a</sup>	87.33 <sup>ab</sup>	354.17 <sup>ab</sup>	812.00 <sup>de</sup>	33.59 abo
Lunadur	V1	4.57 <sup>f</sup>	16.25 <sup>bc</sup>	88.83 <sup>ab</sup>	392.67 <sup>b</sup>	806.83 <sup>cde</sup>	34.55 <sup>cd</sup>
	V2	4.76 <sup>g</sup>	15.40 <sup>a</sup>	88.17 <sup>ab</sup>	344.83 <sup>ab</sup>	815.50 <sup>e</sup>	32.98 <sup>a</sup>
Auradur V	V0	3.35 <sup>d</sup>	15.57 <sup>a</sup>	89.50 <sup>a</sup>	340.83 <sup>ab</sup>	787.67 <sup>cde</sup>	35.44 <sup>d</sup>
	V1	3.54 <sup>ab</sup>	15.75 <sup>ab</sup>	87.50 <sup>ab</sup>	338.33 <sup>a</sup>	784.33 <sup>cd</sup>	35.85 <sup>d</sup>
	V2	3.51 <sup>a</sup>	16.70 <sup>c</sup>	90.00 <sup>a</sup>	351.33 <sup>ab</sup>	770.33 <sup>ab</sup>	37.72 <sup>e</sup>

**Table 5.** Yield components of three durum wheat varieties used in this study. The lower case letters (a, b, c) indicate significant differences among the varieties determined by Tukey's HSD test, included is also standard deviation ( $\pm$ SD). \*\* $\alpha$  = 0.01 indicate level of significance.

	Auradur	Elsadur	Lunadur
Grain yield (GY) (t $ha^{-1}$ ) **	$3.47\pm1.51~\mathrm{a}$	$3.77\pm1.80\mathrm{b}$	$4.59\pm1.74~\mathrm{c}$
Protein content (PC) (%) **	$16.01\pm1.98\mathrm{b}$	$15.54\pm0.57~\mathrm{a}$	$15.73\pm0.80~\mathrm{ab}$
Vitreousness (V) (%) **	$89.00\pm4.65~\mathrm{a}$	$84.67\pm2.70~\mathrm{b}$	$88.11\pm5.10~\mathrm{a}$
Falling number (FN) (s) **	$343.50\pm67.83~\mathrm{ab}$	$329.39 \pm 55.27$ a	$363.89 \pm 48.29  \mathrm{b}$
Bulk density (BD) (g $l^{-1}$ ) **	$780.78 \pm 18.64$ a	$766.50 \pm 26.21$ a	$811.44\pm8.33~b$
Gluten content (GC) (%) **	$36.34\pm4.29~b$	$33.61\pm1.77~\mathrm{a}$	$33.71\pm2.70~\mathrm{a}$

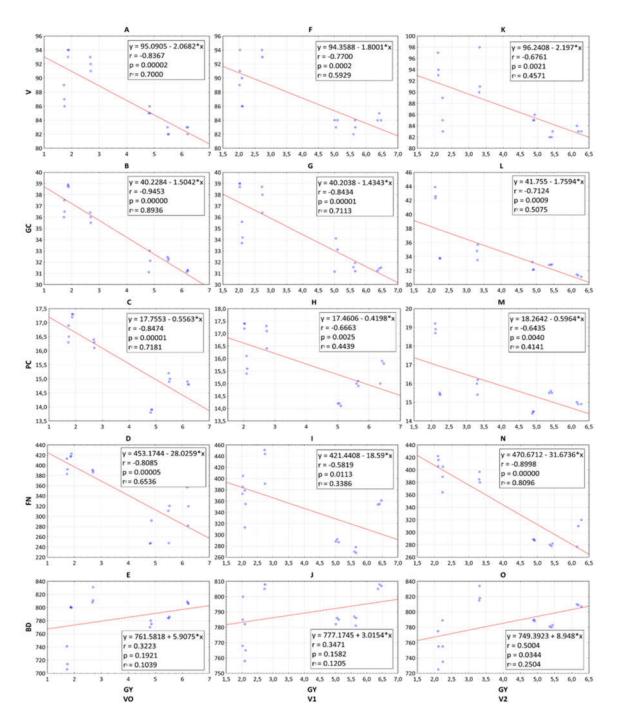
3.3.3. Biofertilization x Variety x Weather Conditions (BVW)

From the ANOVA point of view for three was interactions, a highly significant effect on all production components can be confirmed, with exception of vitreousness and falling number (Table 2). There were significant differences among the individual variations, but as highlighted in Supplementary Table S1, the highest grain yield was achieved in the BVW interaction V1–Lunadur–2018. On the other hand, the highest values of qualitative parameters, except FN and BD, were observed in the interaction V2–Auradur–2017. This finding is particularly interesting because, as mentioned above, the overall impact of biofertilization on production quality was not significant, as well as the BW interaction. In can therefore be assumed that the mentioned interaction V2–Auradur–2017 had the highest influence on the achieved results in relaation to the genetic basis of the variety.

## 3.4. Correlation Analysis for Production Components

Correlation analysis was performed to understand relationships among the monitored components (Figure 3, Supplementary Figures S1 and S2, respectively). In this study, a negative correlation was determined between grain yield and other production components, except bulk density (Figure 3). An important finding was examined in terms of correlations between grain yield and qualitative components on the solved variants in the context of the values from Table 3. They suggest that the bioactive substances did not significantly affect the qualitative components, although as can be seen in Figure 3, the correlation coefficient between GY and quality components (V, GC, PC, FN) decreased on most variants with applied biofertilizers. This suggests that the application of biofertilizers not only led to an increase in grain yield, but also had a positive effect on quality. Correlations between GY and PC, V, FN, and GC were in the range of -0.5819 to -0.9453 (Figure 3A–D,F–I,K–N). The highest negative correlation was found between GY and GC at the level of r = -0.9453 (p < 0.0000) (Figure 3B). On the other hand, the highest positive correlation was observed between GY and BD, at the level of r = 0.5004 (p < 0.0344) (Figure 3O).

In contrast, mutual relationships between baking quality parameters, such as gluten content, vitreousness, protein content, and falling number, were positively correlated in the range of 0.7590–0.9249 (Supplementary Figure S1). The highest positive correlation was observed between PC and GC at the level r = 0.9249 (p < 0.0000) (Supplementary Figure S1). Bulk density of grain did not have a meaningful association with other quality parameters, and correlation values were in the range -0.2681–0.1359 (Supplementary Figure S2).



**Figure 3.** Relationships between grain yield (GY) on all experimental treatment (V0–V2) and vitreousness (V) (**A**,**F**,**K**), gluten content (GC) (**B**,**G**,**L**), protein content (PC) (**C**,**H**,**M**), falling number (FN) (**D**,**I**,**N**) and bulk density (BD) (**E**,**J**,**O**). The linear relationship between parameters is represent by solid lines. Linear equations, correlation coefficient, probability and regression are inserted inside graphs.

# 4. Discussion

This study aimed to evaluate the impact of bioactive substances applied foliarly to durum wheat plants in the field and record changes in quantitative and qualitative characteristics. Especially in dry and hot areas of central Europe, it is currently a significant challenge for farmers to keep crop productivity high due to adverse weather conditions during vegetation. In particular, prolonged drought during critical stages of growth causes a reduction in yields, although the impact on quality is still under discussion. In this experiment, various sources

of bioactive substances in combination with different genetic materials were analyzed and compared for two years to increase grain yield and maintain quality.

The use of bioactive substances in agriculture (syn. bioactive matters, bioactive compounds, biostimulants, etc.) has been reported in many previous studies [18,41,42]. Hence, the application of bioactive substances based on seaweed extract and humic acids was monitored. In both cases, biofertilization was realized three times during the vegetation. It is undisputed that applying these substances has led to an increase in grain yield compared to the non-fertilized variant. However, the best results were obtained on the humic acid variant (Table 3). Ercoli et al. [43] reported an increasing effect of biofertilization with sulfur on grain yield, and similar results were confirmed by Ercoli et al. [44]. The effectiveness of biofertilizer applied alone for improving yield is very poor, but when applied in combination, it may have a beneficial influence [45]. A significant impact of foliar spraying with micronutrients on durum wheat yield components was indicated by Narimani et al. [46]. Likewise, foliar application of seaweed extract had a promotion effect on growth parameters and yield components [47].

In addition to the amount of yield, it is necessary to focus on grain quality. This study identified no statistically significant impact of biofertilization (Table 3) on quality components (gluten content, protein content, vitreousness, falling number, bulk density). However, it is essential to note that, in most cases of the variants with seaweed and humic acids, the quality was maintained or slightly increased. The opposite conclusions were published by Knapowski et al. [48], where applied biofertilizers significantly determined the protein content, falling number, and other baking quality parameters. Differing results are reported in the literature about the impact of biofertilizers on plant yield and quality. Whereas Rašovský and Pačuta [49] found a significant impact on production quantity and non-significant impact of seed-applied biofertilizers on both criteria of common wheat. Likewise, Behera and Rautaray [51] compared recommended doses of chemical fertilizers with biofertilizers applied on durum wheat and found a non-significant impact.

The course of weather conditions had the most significant influence on production components. This is clearly demonstrated by the evaluation of biofertilization x weather conditions interaction (BW). In 2017, a marked lack of precipitation was recorded during the entire growing season (Figure 1), and as a result, significantly lower yields were found compared with 2018 (Table 4). The seasonal water requirement of wheat is represented by the range 450–650 mm [52]. Unfavorable and extreme climate conditions in connection with increased occurrence and impact are considered the greatest danger in wheat production [53]. Nevertheless, the highest grain yield was achieved on BW V1–2018 mainly due to favorable conditions, and the highest increase in grain yield was observed on BW V2–2017 (Table 4). As Van Oosten et al. [54] have already stated, the effects of biostimulants are yield enhancement and resistance to biotic and abiotic stress. Positive effects of seaweed-based biostimulants in drought stress conditions were recorded by Kumar et al. [55] and Goñi et al. [56]. Further studies report an improvement effect of humic substances contained in biostimulants [57,58]. The combination of biostimulant application and drought makes it possible to evaluate the accuracy and effectiveness of the system in research into the effect of biostimulants on drought tolerance [59]. By evaluating the BW interaction in terms of quality, opposing conclusions were found. In most cases (except bulk density), the higher values were observed in 2017 with no statistical differences between variants (Table 4). Fois et al. [60] confirmed the significant negative impact of environmental conditions on yield and quality formation of durum wheat. However, in many previous studies, conflicting information is given in terms of the effect of high temperatures on the quality or protein content of grain [61–63].

One way to increase the effect of bioactive substance application is through the interaction with appropriate genetic material. Beltrano and Ronco [64] suggest that a plant's response to water stress depends on its developmental stage, genotype, and size and duration of the stress. This was confirmed, and high variability in BV interaction for all components was found. Existing wheat genotypes and their uniqueness provide a source of genetic variability, from which many features of interest, e.g., drought tolerance, improvements in nutrient use efficiency, and others, can be selected [65]. We can confirm that different genotypes had highly significant effects on all production components monitored in this experiment. In addition, the highest grain yield and the lowest gluten and protein contents were found in the BV interaction V2–Lunadur. This was not surprising because some previous studies reported similar conclusions [66–68]. Jankowski et al. [69] claim that the influence of different foliar fertilizer applications of FN is dependent on precipitation during the final stages of growth. In this study, minimal differences in vitreousness were found in the BV interaction. Nevertheless, among the analyzed components of wheat cultivars, in the work of Janczak-Pienazek et al. [70], the highest variability was found in grain vitreousness [71]. High variability was not found in the BV interaction for bulk density. As Ložek et al. [72] report, the foliar application of humate has a stimulating effect on yield, but the bulk density of grain was not affected.

A negative relationship between quantitative and qualitative parameters of field crops is well established [73–75] and therefore was expected. Highly significant negative relationships were observed between the grain yield and quality components (except bulk density). Using correlation analysis, Simmonds [76] and Gagliardi et al. [77] wanted to find relationships between quantity and quality parameters of wheat production and observed similar results, mainly for grain yield and protein content. A significant challenge in wheat breeding is a shift in the undesired correlation between these elements [78]. Mutual relationships between almost all quality parameters were in positive correlation dependence. However, Rharrabti et al. [79] reported a general non-significant correlation coefficient.

Despite what has been mentioned above, based on the results of this experiment, it can be confirmed that the Lunadur variety was the most successful in our experimental site, especially in combination with the application of humic acids. On this basis, it is possible to recommend this combination of variety and bioactive substances for practice. A very important component in plant management is the economic efficiency of applied methods [80]. The calculation of economic efficiency was not the aim of this experiment, but it is indisputable that the regular application of biostimulants, compared to conventional technology, is highly dependent on the economy [81]. Calvo et al. [82] confirm that the application of biostimulants increases crop productivity and quality, while responding to economic and sustainable requirements.

## 5. Conclusions

In this study, the impact of various sources of bioactive substances on durum wheat yield and quality parameters was evaluated. Both seaweed- and humic-based preparations were foliarly applied three times during the vegetation, and highly significant differences in grain yield were found in comparison with the control variant. The highest grain yield was achieved on the variant with applied humic acids (V2), although there was no significant difference between V1 and V2. No statistical impact on quality parameters was found, but a positive effect can be concluded in most cases. The different course of weather conditions in the experimental years had the highest impact on all production components. Evaluation of the BW interactions showed that the most significant results were for BD, FN, GC, PC, and V observed in the wheat-friendly conditions of 2017. Significant variability in the results was observed between the durum wheat varieties, which was expected due to their different genetic bases. In general, an important finding was that the highest values of PC, GC, and V were found in the interaction of the Auradur variety on the variant with the applied humic acids.

During the experiment, there was high variability in terms of the influence of factors on the monitored parameters. Ensuring a higher degree of constancy will be a major challenge in the future. In addition, it is necessary to focus on increasing the impact of bioactive substances on the quality parameters of cultivated crops. We recommend the foliar application of bioactive substances in the durum wheat production system primarily because of an important increase in grain yield. However, the impact on quality parameters should be subject to further investigation.

**Supplementary Materials:** The following are available online at https://www.mdpi.com/article/10 .3390/agronomy11071270/s1. Figure S1: Mutual relationships between quality parameters: vitreousness (V), falling number (FN), gluten content (GC) and protein content (PC). Figure S2: Relationships between bulk density (BD) and other quality parameters: vitreousness (V), falling number (FN), gluten content (GC) and protein content (PC). Table S1: Three-way interactions between weather conditions of experimental years, varieties and biofertilization treatments for grain yield (GY), protein content (PC), vitreousness (V), falling number (FN); bulk density (BD) and gluten content (GC).

**Author Contributions:** V.P. prepared the topic of the experiment. M.R. conducted the experiment, realized experimental measurements and statistical analysis in STATISTICA 10 software. B.M.-K. and Z.W. obtained formal analysis, tables and figures. V.P. and M.R. wrote the original manuscript, conducted the investigation and discussions. All authors have read and agreed to the published version of the manuscript.

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