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Plant Growth, Yields and Fruit Quality of Processing Tomato (*Solanum lycopersicon* L.) as Affected by the Combination of Biodegradable Mulching and Digestate

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Abstract: In order to improve environmental sustainability of tomato cultivation and the quality of the harvested fruits, we tested (a) the digestate from anaerobic fermentation of buffalo slurries as partial replacing of NP fertilizers and (b) the biodegradable mulching to improve the nutrients and water availability for crop and to control weeds. In 2017–2018, a private farm of Campania region hosted a trial with four treatments deriving from the combination of two experimental factors: (1) fertilization strategy (standard farm NPK fertilization vs. digestate combined with reduced rates of NP fertilizers); (2) soil mulching (biodegradable mulching vs. no mulching). We measured fresh and dry aboveground biomass (fruits and stem + leaves), yields, fruits quality. Results pointed out: (1) combination of digestate with reduced rates of NP fertilizers did not decrease yields compared to complete mineral fertilization; (2) yields were improved in 2017 by synergic effects of soil mulching and combination of digestate and reduced rates of NP fertilizers; (3) in both the years, digestate combined with reduced rates of NP fertilizers and soil mulching determined the significant improving of fruits quality parameters interesting the processing industry, namely, fruit color, and firmness, total soluble solids, titratable acidity while antioxidant activity, contents of ascorbic acid, polyphenols, flavonoids, and lycopene showed responses variable with year or cultivar.

Keywords: Mater-Bi® based mulch; organic fertilization; tomato yield; antioxidant compounds; total soluble solids; fruit color; fruit firmness

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1. Introduction

Italy is the main producer of processing tomato in the European Union. In 2019, 5.09 million of tons were produced on an area of 77.430 ha [1]. Emilia Romagna and Puglia regions represent the most important productive areas with 26.160 ha and 1.8 million tons the former, with 17.920 ha and 1.6 millions of ton the latter. Campania region is the fourth after Lombardia: 3810 ha are cultivated with a production of 236.000 t concentrated more than 50% in Caserta province. Processing tomato agroecosystems are very intensive, mainly based on monoculture and a large exploitation of chemical fertilizers, pesticides, herbicides, water in order to get high yield enough to allow farmers to gain a profit margin taking in account the low price of tomatoes. N requirements of tomato crop, N above-ground biomass concentration and dilution curves were studied in Central Italy by Tei et al. [2], in Southern Italy by Elia and Conversa [3]. Hartz and Bottoms [4] in California, referred also to the study of Tei et al. [2] to select the best N fertilization practices in order to increase the yields of drip-irrigated processing tomato. All these Authors found that a seasonal N rate of ≈200 kg ha^{-1} appeared adequate to maximize total fruit yield over 120 Mg ha^{-1}.

In the Province of Caserta (Campania region, Southern Italy) there is the highest concentration of the buffalo livestock farming, source of the milk for the productive chain of "mozzarella" cheese. In order to correctly manage the buffalo sludges according to the Action Plan of the Campania Region [5] descending from Nitrate Directive (Council Directive 91/676/EEC), their anaerobic digestion represents a useful piece of a territorial productive system inspired by the principles of circular economy. Biogas and electric power are the main products of an anaerobic digester system, while solid and liquid fractions of the digestate are the by-products. The present research was intended to close the loop to recycle organic carbon and other nutrients by joining the need of the anaerobic digester plant to valorize the digestate with the need of farmers to supply organic fertilizers in the soil. Solid digestate from cattle slurry and manure was tested by Maucieri et al. [6] in a two-year succession of vegetable crops while Nicoletto et al. [7] tested solid digestate from distillery residues in a five-year successions of ten vegetable crops in open field. Both the types of digestates can be defined as organic fertilizers C-N-P-K that in the longer succession showed to efficiently and completely replace mineral fertilizers taking into account the recorded yields and the nitrogen utilization efficiency indexes. Authors recommended only for early spring crops a combination of 50% organic and 50% mineral N, the latter amount to satisfy crop requirements in the initial phase of cycle with lower soil temperatures. Liquid and pelleted digestates from crop biomasses, cow slurry, and grape stalks were used in combination to biochar to feed processing tomato in an organic farm [8]; the highest yields and N agronomic efficiencies were recorded with liquid digestate combined to biochar.

Mulching is a technique mainly used for the production of vegetable crops that influences many factors of the plant-soil-air environment [9]. Conventional mulching is based on non-renewable and not biodegradable materials generating detrimental troublesome residues [10]. The two main advantages of biodegradable films are the adjustment of the films lifetime to that of the agricultural life cycle and their suitability to the prevailing recycling systems in agriculture: in soil biodegradation and on-farm composting [11]. Recent studies are demonstrating the repeated application of different bio-based, biodegradable films incorporated in soil at the end of crop cycle do not modify soil physical properties, soil health indicators and soil functions [12,13]. Paper mulch degrades completely in soil in less than 12 months while Mater-Bi® (NOVAMONT), Ecovio® (BASF) can degrade completely but more slowly depending on climate, soil texture, and biological activity [14,15]. Processing tomato cultivated on Mater-Bi mulch films yielded as much as polyethylene (PE) film [16,17]. Most authors agree that black biodegradable films do not modify the main quality parameters of tomatoes compared to those produced with PE mulch or on bare soil [16–18]. Nonetheless, in our previous studies carried out under tunnel or in open field in South Italy, we have measured, in muskmelon, strawberry, and tomato grown on Mater-Bi films in comparison to PE, significant improvements of fruit quality parameters such as fruit color, total soluble solids, fruit firmness, polyphenols, carotenoids, ascorbic acids, antioxidant activity, etc. [19–22].

Our research was located in private agricultural farms in Villa Literno (Caserta) specialized in processing tomato production with high mechanization. Farmer's fertilization strategy largely exceeds N and P mineral input with rates of 350–400 kg ha^{-1} N and over 300 kg ha^{-1} P. With the aim to improve the sustainability of the agroecosystem, in agreeance with the farmer, an experiment was designed in order to reduce the input of N and P fertilizers by replacing them with solid fraction of digestate from a biogas plant treating buffalo slurries and manure in the neighboring territory. Besides, in order to improve the nutrients flux from soil to plant, control weeds, reduce water irrigation, and to harvest tomato fruits cleaned from soil residues, the adoption of biodegradable, Mater-Bi based mulch (BDM) was tested. Crop responses to the experimental factors was assessed in term of accumulation of dry matter in above-ground biomass, total and marketable yields, fruit quality expressed as mean fruit weight, color, firmness, total soluble solids, dry residue, titratable acidity, antioxidant activity, contents of ascorbic acid, polyphenols, flavonoids, and lycopene.

2. Materials and Methods

2.1. Experimental Site and Design

The research was carried out in 2017 and 2018, in a private, agricultural farm specialized in the tomato processing crop and located in Villa Literno (Caserta province, Campania Region, 41°01′ N and 14°08′ E, 10 m above sea level). Soil chemical characteristics were: pH 7; Electrical Conductivity (1:5 *w/v*) 0.25 mS/cm; Organic carbon 1.25% (=organic matter 2.15%); total Nitrogen 0.18%; available P_2O_5 212.2 ppm; exchangeable K_2O 2451 ppm. It has to be pointed out the high contents of phosphorus (probably consequence of exceeding fertilization through the years) and potassium (native property of soil). Two experimental factors, each in two levels, were studied: (1) plant fertilization managed according to the conventional, complete, mineral fertilization applied in the farm (Control) or according to an integrated approach with solid fraction of digestate from buffalo slurries and manure combined with a reduced rates of N-P mineral fertilizers (INT); (2) soil mulching articulated in two levels: bare soil usually practiced in the farm (Bare) and soil mulched with a compostable/biodegradable film (BDM). The treatments were laid out according to a strip-split plot design with ten replications. Either in 2017 or in 2018, the same distribution of the four treatments was applied in the same area of 5200 m^2 devoted to the experiment in the private farm. Due to the farmer's requirement for a simple design, manageable with farm machineries, strip main plots hosting the two fertilization treatments were disposed of one adjacent to the other in soil rectangles of 180 m × 14.4 m each. Soil mulching was split inside each main plot forming two adjacent sub-plots of 180 m × 7.2 m. In the plots hosting each factorial treatment, ten replications constituted by groups of eight plants were randomly individuated.

Table 1 represents the schemes of fertilization followed to manage the Control and the Integrated treatments.

Table 1. Total amounts of N-P_2O_5-K_2O supplied to tomato in pre-transplant and post-transplant in the Control and Integrated scheme of fertilization put in practice in 2017 and 2018.

Fertilization *	Pre-Transplant		Post-Transplant via Fertigation		Total Amount of Nutrients
	Type of Fertilizers	**Nutrients ($Kg\ ha^{-1}$)**	**Type of Fertilizers**	**Nutrients ($Kg\ ha^{-1}$)**	**($Kg\ ha^{-1}$)**
Control	Diammonium phosphate. Nitrophoska blu (12-12-17), Organosprint (12-15-5)	N: 140 P_2O_5: 240 K_2O: 60	Different commercial fertilizers split into 15 applications	N: 220 P_2O_5: 140 K_2O: 120	N: 360 P_2O_5: 380 K_2O: 180
INT	Digestate solid fraction (50–40 Mg ha^{-1} as fresh weight per each year) **	N: 150 P_2O_5: 70	Different commercial fertilizers split into 15 applications	N: 150 P_2O_5: 175 K_2O: 170	N: 300 P_2O_5: 245 K_2O: 170

Legend: * Control is the standard fertilization applied by farmers, INT is the integrated fertilization with digestate and reduced rates of N-P fertilizers; ** these amounts contained 150 kg ha^{-1} of total N to comply with Action Plan of Campania Region [5] following to the Nitrate Directive (Council Directive 91/676/EEC).

INT treatment was intended to start a shift of farm practice toward a more sustainable fertilization management. In this step, we slightly reduced the full fertilization formula by cutting 60 and 135 kg ha^{-1} of N and P_2O_5 with respect to Control treatment. Instead, a significant cut was represented by the saving of NP mineral fertilizers: 210 kg N ha^{-1} (150 from digestate + 60 as savings of N mineral fertilizers) and 205 kg ha^{-1} of P (in average 70 from digestate + 135 as savings of P min fertilizers).

Solid digestate was supplied by Power Rinasce S.p.A. anaerobic digester plant of S. Maria La Fossa (CE). After separation from liquid phase, solid digestate was kept on concrete ground for three months. Its main chemical characteristics in 2017 and 2018 are shown in Table 2.

Table 2. Main chemical characteristics of digestates utilized in the two years of the experiment.

Parameters	Units	2017	2018
pH		8.1	8.1
Dry matter (d.m.)	%	45	49
Organic C	% d.m.	24.8	36.6
Total N	% d.m.	0.66	0.76
N-NH_3	% d.m.	0.08	0.10
Organic N	% of total N	88	87
P_2O_5 *	% d.m.	0.3	0.37
Electrical Conductivity	mS cm^{-1}	0.55	0.53

* Phosphorus pentoxide (P_2O_5 expressed as ortophosphate PO_4 is 0.4% in 2017 and 0.5% in 2018).

Biodegradable mulch treatment was a Mater-Bi®-based, grade EF04P, black, 15 µm thick, 120 cm wide film of Novamont S.p.A. Other technical data regarding physical and mechanical properties for this grade and thickness of Mater-Bi are: Tensile strength 35 MPa, Elongation at break 380%, Young modulus 200 MPa, Density 1.27 g cm^{-3}. Biodegradable mulch was preferred to LDPE mulch in order to exploit the soil mulch advantages [9] but avoiding (a) the costs for collecting and disposing of polyethylene mulch after the end of crop cycle and (b) solving the problem posed by polyethylene mulch films during mechanical harvesting when the film is ripped and dispersed on the soil.

Bare soil was the standard practice applied in the farm where control of weeds was usually chemical and mechanical.

2.2. Crop Cycle

Tomato transplant occurred on 27 April 2017 and 24 April 2018; plantlets, in the first year, were planted by a transplanter in double-rows bed in bare soil; on the contrary, plants were hand-planted in mulch soil. Rows on the bed were 0.5 m apart, plantlets on the rows were 0.4 m apart, while distance between the middle of two adjacent beds was 1.2 m. Therefore, plant density was 33.000 plants ha^{-1}. In the second year, farmer used a transplanter combined to a mulch system layer in order to completely mechanize the operations. In the first year was cropped HF1 5508 (HeinzSeed, Stockton, CA, USA) while in the second year was cropped HF1 Kendras (Nunhems Italy, BASF, S.Agata Bolognese, Italy) with the aim to test the quantitative and qualitative responses of different cultivars to the same experimental design. Both the cultivars produced round shape fruits. Drip irrigation and plant protection from pests were managed according to the farm standard.

Harvesting in the replication areas was hand-made and occurred on 19 August 2017 and on 9 August 2018.

2.3. Growth and Yield Parameters

In order to assess the accumulation of fresh and dry matter in stems + leaves and fruits of tomato, during the crop cycle were sampled 5 plants in pre-selected positions external to the replicates in each of the four treatments. Times of sampling were at 3, 7, 12, 16 (in 2017) or 15 (in 2018) weeks from the day of transplant. After the measure of the fresh weight, the aboveground biomass was dried in oven at 70 °C and then weighted. When more than 80% of fruits were ripened, in each replicate with 8 plants, fruits were collected and graded in red, green, pink, and not marketable. Mean fruit weight was assessed by weighting a sample of 15 ripen fruits. After the hand-made harvest of the ten replicates, the mechanical harvesting of the whole experimental area was affected so that it was possible to evaluate the behavior of BDM.

2.4. Fruit Quality

At harvest 10 ripen fruits were collected from each replicate to determine dry residue, soluble solids, color and firmness, pH and total titratable acidity. In addition, another 10 ripen fruits were collected from 3 out of 10 replicates, washed with ultrapure water, dried and stored in the freezer (−80 °C) to perform the following analyses: ascorbic acid, lycopene, total phenols, total flavonoids, and antioxidant activity.

2.5. Chemicals and Instruments

All reagents and standards were of analytical reagent (AR) grade. L-ascorbic acid (L-AA) (>99.8%), Folin–Ciocalteu's phenol reagent 2N solution (for protein analysis), 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox) (>98%), 2,2-diphenyl-1-picrylhydrazyl (DPPH) (>95%), (+) catechin hydrated (>99%), metaphosphoric acid (MPA) (33.5–36.5%), gallic acid (>98%), sodium hydroxide (>98%) acetic acid (>99%), ethylenediaminetetraacetic acid disodium salt (EDTA) (98%) were obtained from Sigma-Aldrich MERCK KGaA affilate (Darmstadt, Germany); sodium nitrite (>97%) were obtained from Honeywell Fluka (Seelze, Germany); sodium carbonate anhydrous (RPE), methanol (HPLC grade > 99.9%), n-hexane (HPLC grade > 99.9%), acetone (HPLC grade) were obtained from Carlo Erba Reagents (Cornaredo (MI), Italy), aluminum chloride hexahydrated (99%) were obtained from Titolchimica S.p.A (Pontecchio Polesine (RO), Italy), Butylated Hydroxytoluene (BHT) (99%) were obtained from Alfa Aesar Thermo Fisher (Kandel, Germany). The colorimetric analysis was performed using a UV-Vis Varian Cary 100 Spectrophotometer (Varian Agilent, Santa Clara, CA, USA). Ultrapure water was supplied ion exchange system, Milli-Q, fed by the reverse osmosis system, Elix 3, both from Millipore (Merck group, Darmstadt, Germany).

2.6. Determination of Physico-Chemical Parameters

2.6.1. Color

Fruit color was measured using a colorimeter (CR-400 Minolta, Osaka, Japan) based on the CIELAB color space represented by L* (brightness), a* (redness), b* (yellowness) values. Measurements were taken at two different points on equatorial area of each fruit.

2.6.2. Firmness

The measurement was performed on the two side at the equatorial zone of the fruit by means of digital penetrometer (T.R. Turoni s.r.l., Forlì, Italy) with a 8 mm tip. The force applied until the 4 mm penetration of the strut was expressed in Newton (N).

2.6.3. Total Soluble Solids (TSS)

The total soluble solids content (°Brix) was measured by DBR 35 portable digital refractometer (Sinergica Soluzioni s.r.l., Pescara, Italy). The measurements were carried out by placing a fruit juice drop in the screen of the refractometer to further assessing the result.

2.6.4. pH and Total Titrable Acidity (TTA)

The acidity (pH value) was measured with a pH meter (Multi-parameter portable meter MultiLine®®F WTW, Weilheim, Germany), equipped with WTW—Low-Maintenance SenTix®® 41 pH electrode and temperature sensor. TTA was measured by titration with NaOH 0.1 M according to official method AOAC [23]. Results were reported as percentage of citric acid.

2.7. Evaluation of Antioxidant Compounds and Antioxidant Activity

2.7.1. Sample Preparation for Chemical Determinations

Samples consisting of 10 tomato fruits were washed with ultrapure water, dried and stored in freezer (−80 °C). Still frozen fruits were roughly cut with a ceramic blade knife, brought back to −20 °C, and homogenized in a knife mill Grindomix GM300 Retsch (Haan, Germany). The grinding program was in two steps: 1000 rpm revolution speed, direction impact, for 20 s (pre-grinding) and 2000 rpm revolution speed, direction cut, for 30 s (fine-grinding). The homogenated obtained were divided into sub-samples and stored at −80 °C and used for the determination of ascorbic acid, lycopene, total phenols, flavonoids, and antioxidant activity.

2.7.2. Ascorbic Acid

Ascorbic acid extraction procedure (in duplicate for each sample) was carried out according to AOAC Official method [24]. Briefly, 10 mL of extraction solution (30 g L^{-1} MPA—80 mL L^{-1} acetic acid—1 mmol L^{-1} EDTA) was added to 3–4 g of homogenate and the mixture was homogenized for 3′ with a disperser (Ultra-TuraxT25 IKA-Werke GmbH & Co. KG, Staufen, Germany). The resulting extract after centrifugation (10.000 rpm; 20 min; 4 °C) was immediately analyzed by colorimetric method using Folin Phenol Reagent according to Jagota and Dani [25]. To 400 µL of sample was added 200 µL of extraction solution, 1.2 mL of ultrapure water and 200 µL of Folin–Ciocalteu's phenol reagent 2N (diluted 1:5 with ultrapure water). After 30 min in darkness and room temperature, the adsorbance of blue color developed was measured at 760 nm. This procedure was repeated in triplicate. For quantification, six standard solutions of ascorbic acid (range 25–150 µg/mL) were prepared and estimated as above ($R^2 \geq 0.996$).

2.7.3. Lycopene

Lycopene, the major carotenoid in tomatoes, was extracted according to the method described by Periago et al. [26]. Approximately 5–6 g of sample (in triplicate) mixed with 25 mL of extraction solvent (hexane/acetone containing 0.5% butylated Hydroxytoluene (BHT)/methanol 2/1/1 by vol.) were magnetically stirred during 30 min in dark glass bottle and at room temperature to avoid lycopene loss by photo-oxidation. The upper organic phase (hexane) was recovered and transferred to a dark glass, separating funnel by filtering on fritted glass funnel. These operations were repeated until the pulp was completely colorless (4–5 times) adding fresh mixture each time. At the end of the extraction cycles, everything was filtered on fritted glass funnel in the same collection funnel of the various extraction fractions. The aqueous phase was removed while the organic phase was transferred to a flask and brought to the final volume of 50 mL with hexane. A 503 nm spectrophotometer reading was carried out on suitably diluted extracts and the lycopene concentration was calculated by Lambert-Beer law (ε_M = 17.2 × 104 M^{-1} cm^{-1}). Lycopene content was expressed in mg kg $^{-1}$ of fresh weight.

2.7.4. In Vitro Chemical Screening of Total Phenols

Extraction was done following the procedure as described by Kaur et al. [27] with minor modifications. Approximately 5–6 g of sample (in triplicate) were homogenized in 25 mL of 80:20 CH_3OH/H_2O mixture and sonicated in an ultrasonic bath (Elmasonic P Elma, Singen, Germany) at 40 °C for 3h into darkness. Afterwards, the extracts were centrifuged for 20 min at 10,000 rpm (HF 14.94 rotor, radius 9.7 1086 RCF) at 4 °C (Contifuge17R Heraeus Sepatech, Waltham, Massachusetts, US) and the upper layer was separated and kept at −20 °C until analyses were performed. The total phenolic content was determined according to Folin–Ciocalteu's method [28] with small modifications. In the tube test were in sequence added 1 mL H_2O, 100 µL of tomato extract and 100 µL of Folin–Ciocalteu's phenol reagent 2N solution and, after 10 min, 800 µL of Na_2CO_3 75 g/L solution so that the final pH was ≥10 (test final volume 2 mL). The mixture was allowed to stand for 120 min at room temperature and into darkness and absorption was measured at 765 nm against a reagent blank. Measurements were performed in duplicate and results were expressed as mg of gallic acid equivalent per 100 mg of fresh weight (GAE 100 g^{-1} fw). For calibration, six standard solutions of gallic acid (range 0–150 µg/mL) were prepared and estimated as above ($R^2 \geq 0.998$).

2.7.5. In Vitro Chemical Screening of Total Flavonoids

Total flavonoid content was determined on tomato extract prepared as above, using the spectrophotometric assay, based on aluminum chloride complex formation, as described by Zhishen et al. [29], with modifications [30]. A known volume of tomato extract was placed in a 10 mL volumetric flask and ultrapure water was added to make 5 mL. Therefore, the following were added in the order: 0.3 mL of $NaNO_2$ (5% *w/v*), 3 mL $AlCl_3$ (10% *w/v*)

5 min later and after 6 min 2 mL di NaOH 1 mol L^{-1}. The mixture was brought to a final volume of 10 mL with ultrapure water and left at room temperature for 15 min. Then the adsorbance was measured at 510 nm. The analyses were carried out in triplicate and the results were expressed as mg catechine equivalent per 100 g of fresh weight (mg CE 100 g^{-1} fw). For quantification, twelve standard solutions of catechine (range 0.25–10 µg/mL) were prepared and estimated as above ($R^2 \geq 0.9998$).

2.7.6. In Vitro Chemical Screening of Antioxidant Activity

The antioxidant activity was measured on tomato extract prepared as for total phenolic determination, using the DPPH assay based on the measurement of the scavenging ability of antioxidant toward the stable radical DPPH [31]. A 3.9 mL aliquot of a 0.0634 mM of DPPH solution in methanol was added to 0.1 mL of each extract and shaken vigorously. Change in the absorbance of the mixture at 515 nm was observed and the antiradical activity of sample was evaluated from the percentage DPPH remaining when the kinetics reached a steady state. It has been found experimentally that for tomato fruit, this occurs after 150 min. Some precautions were taken because the stability of the radical DPPH is dependent on exposure to light and oxygen content [32]. A stock solution of DPPH 634 µM (12.5 mg in 50 mL of degassed methanol) was prepared at the assay moment. For the test, instead, a 10-fold fresh diluted solution is used. Dark glass test tubes were used with teflon-coated rubber septa and aluminum caps (Supelco Inc., Bellefonte, PA, USA), and the DPPH solution was degassed and flushed with helium gas for 3 min. Trolox equivalency is used as a benchmark for the oxidant capacity of the extracts. Therefore, the calibration curve was constructed using nine standard solutions containing this antioxidant (range 0.15–20 µM) and the results are expressed as µmol of Trolox equivalent per 100 g of fresh weight (µmol TE 100 g^{-1} fw).

2.8. Statistical Analysis

Data collected in the trials of 2017 and 2018 were separately analyzed due to the different tomato cultivar cropped; as a result, the effect of year and cultivar overlap and confuse. Per each year, all agronomical and qualitative data were subjected to Two-way analysis of variance (ANOVA) for two main experimental factors, 'Fertilization strategy', 'Soil mulching', and their interaction. When appropriate, mean separations were performed through Tukey's HSD test to 0.05 probability level using the Statistica software (1996).

3. Results

3.1. Processing Tomato Growth

In Table 3 are showed minimum and maximum temperature, rain measured during the months of tomato cultivation. In 2017 and 2018, both minimum and maximum temperatures showed a similar trend. Besides, the rainfall was low in the years of the trial with the only exception of May 2018 when it rained 104 mm.

Figure 1 shows the effects of the different combinations of the experimental factors on the trend of dry matter accumulation in stems plus leaves along the crop cycle in 2017 whereas, Figure 2 shows the trend of dry matter accumulation in fruits.

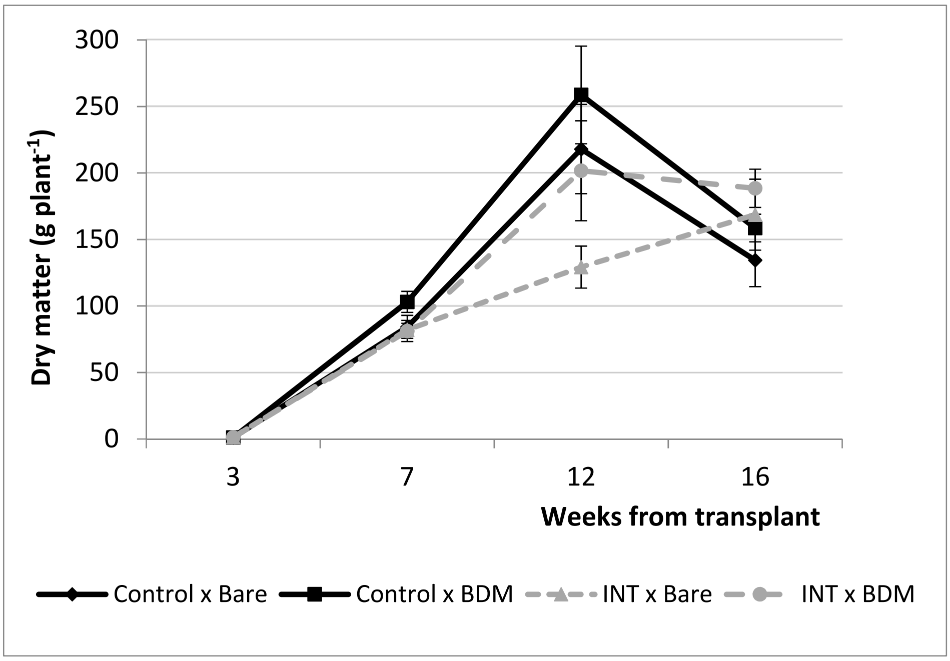

Figure 1. Dry matter accumulation in stems and leaves biomass of tomato cv Heinz 5508 during the 2017 crop cycle.Legend: vertical bars represent error standard of mean (N = 10) per each combination of treatments in different sampling times. Control × Bare: standard farm fertilization × Bare soil; Control × BDM: standard farm fertilization × Biodegradable mulch; INT × Bare: solid digestate integrated with mineral fertilizers × Bare soil; INT × BDM: solid digestate integrated with mineral fertilizers × Biodegradable mulch.

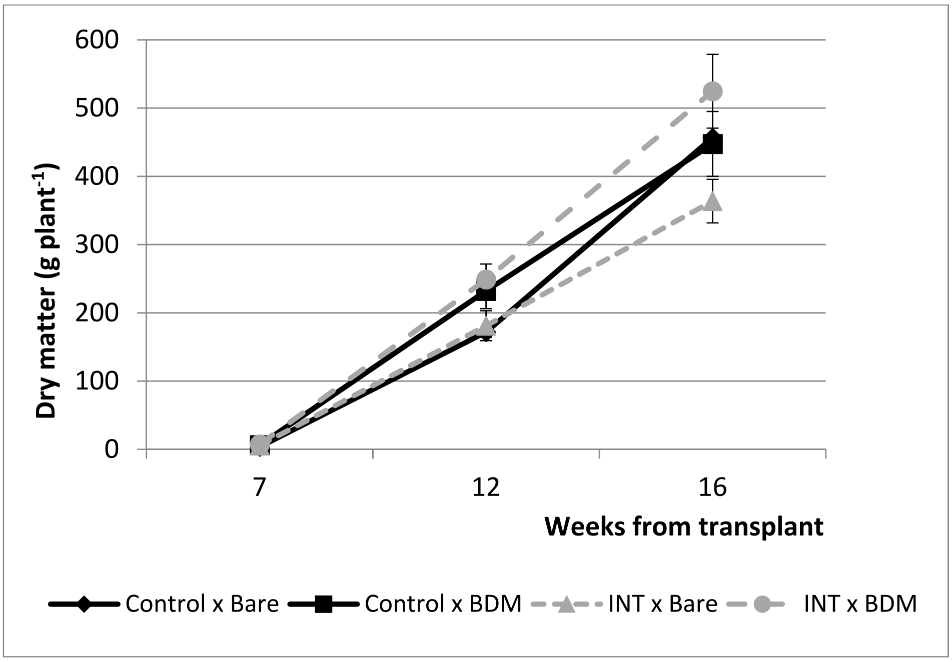

Figure 2. Dry matter accumulation in fruits of tomato cv Heinz 5508 during the 2017 crop cycle. Legend: vertical bars represent error standard of mean (N = 10) per each combination treatments in different sampling times. Control × Bare: standard farm fertilization × Bare soil; Control × BDM: standard farm fertilization × Biodegradable mulch; INT × Bare: solid digestate integrated with mineral fertilizers × Bare soil; INT × BDM: solid digestate integrated with mineral fertilizers × Biodegradable mulch.

Table 3. Minimum and maximum temperature (°C), cumulated rain expressed as monthly means from April to August 2017 and 2018 in Grazzanise (Caserta).

2017	April	May	June	July	August
Tmin °C	8.2	12.7	19.5	19.6	20.3
Tmax °C	19.7	25.2	30	31.8	33.5
Rain (mm)	7.4	7.2	7	16.8	0
2018					
Tmin °C	11.4	14.9	18.3	20.9	21.9
Tmax °C	22.7	24.6	29.3	31.7	33.2
Rain (mm)	0	104.4	14	11.4	10.2

Accumulation of dry biomass in stems and leaves was not influenced significantly by fertilization, mulch or their interaction with the exception of the mean effect of fertilization on week 12 from transplanting when, in average, both the treatments with standard farm mineral fertilization (Control) increased the amount of dry matter per plant in comparison to the digestate integrated with chemical fertilizers (INT). Maximum accumulation of dry matter was recorded at week 12 with 200–250 g plant^{-1} in all treatments, excluding the INT × Bare combination (130 g plant^{-1}). In this phase, it was evident the strong vegetative boost determined by the Control fertilization; indeed, the relative growth rate between weeks 7 and 12 was, in average, 4.7 g day^{-1} in Control treatments against 2.7 g day^{-1} in INT ones (data not showed). With the increasing of fruit set and development, a large translocation of dry matter from leaves and stems to fruits explain the fall of its content recorded on week 16 at harvest, particularly in the Control treatments.

At the same time, starting from week 7, it is observed a continuously growing trend of dry matter accumulation in fruits until the harvest (Figure 2). Biodegradable mulch influenced significantly and positively a major accumulation both in Control or INT fertilized plots on weeks 7 and 12 while at the harvest time (week 16), the highest mean dry matter accumulation of 524 g plant^{-1} was measured in INT × BDM, about 450 g plant^{-1} in Control combined to BDM or Bare, the lowest accumulation of 363 g plant^{-1} in INT × Bare.

Figures 3 and 4 show the dry matter accumulation recorded, respectively, in stems and leaves or in fruits during crop cycle in 2018. Dry matter accumulation follows the same pattern as in the preceding year as concerns stems and leaves.

On week 3 significant mean effects of Fertilization and Mulch were detected; young plantlets showed more dry matter in stems and leaves when they grew in Control and INT plots not combined with biodegradable mulch film. No significant effects were attributable to the experimental factors in any successive week of sampling.

As in the 2017 crop cycle, the peak of the vegetative development was on week 12 and the relative growth rate calculated with respect to the week 7 was on average 6.1 g plant^{-1} in Control treatments against 2.5 g day^{-1} in INT ones (data not showed). At the harvest, dry matter in stems and leaves fell as expected. On the opposite, trend of dry matter accumulation in fruits was irregular (Figure 4). Looking at the accumulation of dry matter in fruits, the ANOVA put in evidence the highly significant, negative effect of mulch at week 7 whit a reduction in fruit settings in comparison to bare soil as a consequence of the delay in growth of plants on BDM as described in week 3 for stems and leaves. Until to week 12, we detected a regular growing accumulation of dry matter also higher than that one measured in 2017: all the treatments on average accumulated 295 g plant^{-1} in 2018 against 208 g plant^{-1} in 2017. At the harvest time, however, dry matter in fruits fell under the preceding level in all the treatments.

At week 15, the significant effect of mulch pointed out a turning of the accumulation in fruits of plant grown on mulch that showed a complete recovery of the initial difficulties.

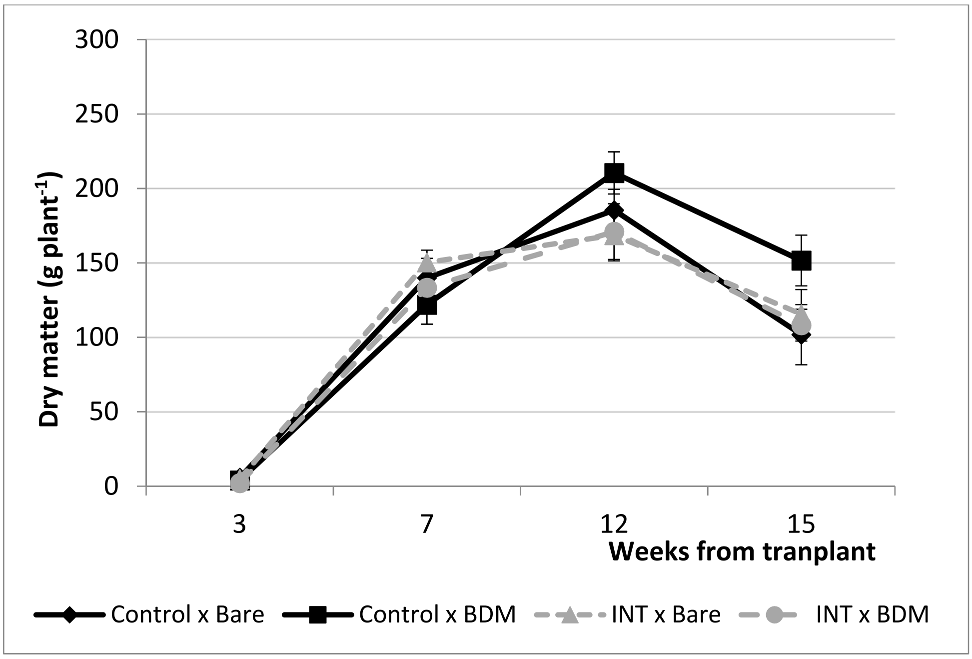

Figure 3. Dry matter accumulation in stems and leaves biomass of tomato cv Kendras during the 2018 crop cycle. Legend: vertical bars represent error standard of mean (N = 10) per each combination of treatments in different sampling times. Control × Bare: standard farm fertilization × Bare soil; Control × BDM: standard farm fertilization × Biodegradable mulch; INT × Bare: solid digestate integrated with mineral fertilizers × Bare soil; INT × BDM: solid digestate integrated with mineral fertilizers × Biodegradable mulch.

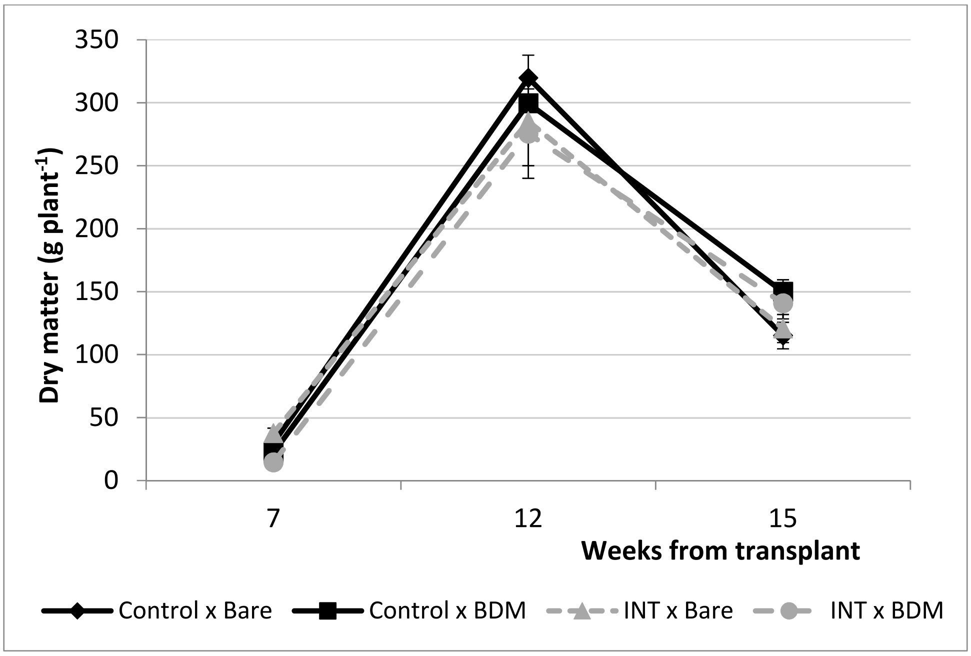

Figure 4. Dry matter accumulation in fruits biomass of tomato cv Kendras during the 2018 crop cycle. Legend: vertical bars represent error standard of mean (N = 10) per each combination of treatments in different sampling times. Control × Bare: standard farm fertilization × Bare soil; Control × BDM: standard farm fertilization × Biodegradable mulch; INT × Bare: solid digestate integrated with mineral fertilizers × Bare soil; INT × BDM: solid digestate integrated with mineral fertilizers × Biodegradable mulch.

3.2. Tomato Yields

In 2017, from June to the harvest occurring in August, the weather was dry and warm, but farmer's management of drip irrigation avoided crop stress. In Table 4, the yields and mean fruit weight of tomato cv Heinz 5508 are shown.

Table 4. Total yield and its composition, fruit mean weight of tomato cv Heinz 5508, in 2017, in relation to different combinations of type of fertilization and soil mulching.

	Marketable Yield	Green Yield	Pink Yield	Not Marketable Yield	Total Yield	Fruit Mean Weight
Treatments			(Mg ha^{-1})			(g)
INT × BDM	154.1 a	7.1 a	8.5 a	3.8 n.s.	173.5 a	73 a
INT × Bare	114.1 c	4.6 ab	5.5 ab	6.0	130.2 c	68 b
Control × BDM	139.6 ab	2.4 b	3.9 b	9.5	155.5 ab	67 b
Control × Bare	123.8 bc	6.4 ab	8.4 a	7.5	146.2 bc	70 ab
Significance						
Fertilization	n.s	n.s.	n.s.	*	n.s.	n.s.
Mulch	*	n.s.	n.s.	n.s.	*	n.s.
Fertiliz. × Mulch	*	*	*	n.s.	*	*

n.s.: not significant differences; * significant differences to Fisher's Test at $p < 0.05$; Different letters within each column indicate significant differences according to Tukey HSD Test ($p = 0.05$). INT = integration of solid digestate and mineral fertilizers; Control = farm mineral fertilization; BDM = biodegradable mulch; Bare = bare soil.

Fertilization effect was not significant on marketable, green, pink and total yield as well as mean fruit weight. Only non-marketable yield was significantly higher in Control (8.5 Mg ha^{-1}) vs. INT (4.9 Mg ha^{-1}). Soil mulching, instead, significantly influenced marketable and total yields that were higher in mulch than bare soil. The other components of the production were not influenced by the kind of soil covering. The interaction Fertilization × Mulch was significant in all the variables excepted for the discarded yield. BDM in combination either with INT or Control fertilizations favored the higher yields, while INT × Bare soil determined the lowest yield although not significantly different from the standard farm practice (Control × Bare). In particular, the combination of fertilization with digestate and soil mulching produced significantly more than the standard farm practice. The not fully ripen part of the production (green and pink) was significantly lower in Control × BDM while in the other treatments ranged between 10–15 Mg ha^{-1}. Mean fruit weight was significantly higher in INT × BDM compared to Control × BDM and INT × Bare.

The yields and mean fruit weight of tomato cv Kendras cultivated in 2018 are shown in Table 5.

Table 5. Total yield and its composition, fruit mean weight of tomato cv Kendras, in 2018, expressed as mean effect of Fertilization and Mulch.

Treatments	Marketable Yield	Green Yield	Not Marketable Yield	Total Yield	Fruit Mean Weight
			(Mg ha^{-1})		(g)
Fertilization					
INT	72.6	4.8	2.0	79.5	71 b
Control	66.0	5.1	2.0	73.1	80 a
Mulch					
BDM	72.0	4.7	2.2	79.7	75
Bare	66.6	5.5	1.9	73.0	76
Significance					
Fertilization	n.s.	n.s.	n.s.	n.s.	*
Mulch	n.s.	n.s.	n.s.	n.s.	n.s.
Fertiliz. × Mulch	n.s.	n.s.	n.s.	n.s.	n.s.

n.s.: not significant differences; * significant differences to Fisher's Test at $p < 0.05$; different letters within each column indicate significant differences according to Tukey HSD Test ($p = 0.05$). INT = integration of solid digestate and mineral fertilizers; Control = farm mineral fertilization; BDM = biodegradable mulch; Bare = bare soil.

The irregular trend of dry matter accumulation in fruits with the fall at harvest time shown in Figure 4, finds confirmation by examining the level of yields recorded in 2018 with cv Kendras. In general, ANOVA did not put in evidence any statistical difference among the treatments. Total yields were generally 50% lower than those ones recorded in 2017. As in 2017, the Fertilization effect was not significant in all the variables; only fruit mean weight in Control was significantly higher than INT. Mulch effect was also not significant, as well as the interaction of the two experimental factors. We point out that the pink-colored fruits were not found at harvest. Confirming the ranking recorded in 2017, marketable and total yields in INT × BDM were 16% higher than in Control × Bare that showed the lowest production.

3.3. Tomato Quality

In Table 6 are shown the physico-chemical parameters of the fruits of tomato cv Heinz 5508, in 2017. Fertilization effect was significant on color components, firmness, total soluble solids, and titratable acidity that were higher in INT than Control. Only pH mean values was significantly higher in Control than INT. Dry residue was not influenced by fertilization.

Table 6. Peel color, firmness, total soluble solids, dry residue, pH, and titratable acidity of tomato fruits of cv Heinz 5508, as influenced by the effects of fertilization and mulch in 2017.

Treatments	L	Color a	b	Firmness (N)	TSS °Brix	DR (%)	pH	TA (% f.w.)
Fertilization								
INT	39.33 a	38.08 a	26.48 a	0.92 a	5.05 a	6.13	4.31 b	0.43 a
Control	33.38 b	29.48 b	19.33 b	0.85 b	4.81b	5.98	4.45 a	0.37 b
Mulch								
BDM	37.31 a	35.63 a	24.07 a	0.90 a	5.05 a	6.06	4.35 b	0.43 a
Bare	35.40 b	31.93 b	21.74 b	0.87 b	4.81 b	6.05	4.41 a	0.37 b
Significance								
Fertilization	*	*	*	*	*	n.s.	*	*
Mulch	*	*	*	*	*	n.s.	*	*
Fertiliz. × Mulch	n.s.	n.s.	n.s.	*	n.s.	n.s.	n.s.	*

Legend: TSS-total soluble solids; DR—dry residues; TA—titratable acidity; n.s.: not significant differences; * significant differences to Fisher's Test at $p < 0.05$; different letters within each column indicate significant differences according to Tukey HSD Test ($p = 0.05$). INT = integration of solid digestate and mineral fertilizers; Control = farm mineral fertilization; BDM = biodegradable mulch; Bare = bare soil.

Soil mulching significantly influenced all physico-chemical parameters with mean values higher in mulch than bare soil, excepted for the pH and dry residue. The pH was significantly higher in bare soil than in mulch while dry residue was not influenced by mulch.

The interaction Fertilization × Mulch was significant on firmness and titratable acidity whose values were significantly improved by the combination of fertilization with digestate and soil mulching in comparison to the other treatments (Figure 5).

The effect of treatments on antioxidant compounds and antioxidant activity assessed in tomato cv Heinz 5508, in 2017 is reported in Table 7. The interaction Fertilization × Mulch was significant on ascorbic acid, polyphenols content and antioxidant activity. For each of these characteristics, the combination Control × Bare soil produced the lowest mean value while the introduction of biodegradable mulch and the integration of solid digestate with reduced doses of N-P fertilizers improved particularly polyphenols and antioxidant activity of fruits. The biodegradable mulch increased lycopene content to 71.8 against 65.9 mg kg^{-1} f.w. in fruits grown on Bare soil.

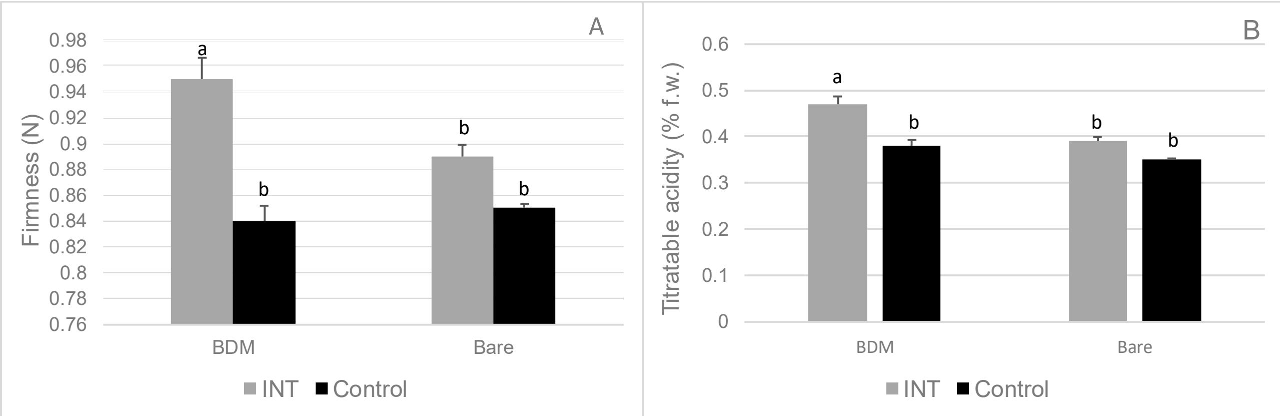

Figure 5. Effects of the interaction Fertilization * Mulching on the firmness (**A**) and titratable acidity (**B**) of fruits of Heinz 5508 in 2017. Legend: vertical bars represent standard error of mean (N = 10) per each treatment. Letters on the top of columns indicate means are significantly different to Tukey HSD Test ($p = 0.01$). Control × Bare: standard farm fertilization × Bare soil; Control × BDM: standard farm fertilization × Biodegradable mulch; INT × Bare: solid digestate integrated with mineral fertilizers × Bare soil; INT × BDM: solid digestate integrated with mineral fertilizers × Biodegradable mulch.

Table 7. Antioxidant compounds and antioxidant activity in tomato cv Heinz 5588, as influenced by the interaction of Fertilization and mulch in 2017.

Treatments	Ascorbic Acid (mg 100 g^{-1} f.w.)	Polyphenols (mg GAE 100 g^{-1} f.w.)	Flavonoids (mg CE 100 g^{-1} f.w.)	Antioxidant Activity (mg TE 100 g^{-1} f.w.)	Lycopene (mg kg^{-1} f.w.)
INT × BDM	19.0 ab	40.2 a	4.63	199.3 a	70.9
INT × Bare	20.6 ab	40.2 a	4.98	208.6 a	65.1
Control × BDM	21.0 a	40.8 a	4.79	197.1 a	72.7
Control × Bare	17.6 b	36.2 b	4.54	166.9 b	66.7
Significance					
Fertilization	n.s.	*	n.s.	*	n.s.
Mulch	n.s.	*	n.s.	*	*
Fertiliz. × Mulch	*	*	n.s.	*	n.s.

n.s.: not significant differences; * significant differences to Fisher's Test at $p < 0.05$; different letters within each column indicate significant differences according to Tukey HSD Test ($p = 0.05$). INT = integration of solid digestate and mineral fertilizers; Control = farm mineral fertilization; BDM = biodegradable mulch; Bare = bare soil.

In Table 8 are shown the physico-chemical parameters of tomato fruits of cv Kendras cultivated in 2018.

Table 8. Peel color, firmness, total soluble solids, dry residue, pH, and titratable acidity of tomato cv Kendras, in 2018, as influenced by the effects of fertilization and mulch.

Treatments	L	Color a	b	Firmness (N)	TSS (°Brix)	DR (%)	pH	TA (% f.w.)
Fertilization								
INT	38.46 a	41.43 a	27.18 a	1.00 a	5.30	5.55	4.39	0.40
Control	35.46 b	37.51 b	25.43 b	0.85 b	5.10	5.60	4.43	0.41
Mulch								
BDM	38.66 a	41.83 a	28.06 a	1.00 a	5.43 a	5.81	4.38	0.40
Bare	35.26 b	37.11 b	24.85 b	0.85 b	4.97 b	5.34	4.44	0.40
Significance								
Fertilization	*	*	*	*	*	n.s.	n.s.	n.s.
Mulch	*	*	*	*	*	n.s.	n.s.	n.s.
Fertiliz. × Mulch	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.

Legend: TSS-total soluble solids; DR—dry residues; TA—titratable acidity; n.s.: not significant differences; * significant differences to Fisher's Test at $p < 0.05$; different letters within each column indicate significant differences according to Tukey HSD Test ($p = 0.05$). INT = integration of solid digestate and mineral fertilizers; Control = farm mineral fertilization; BDM = biodegradable mulch; Bare = bare soil.

Fertilization, as well as Mulch effects, were significant on color components, firmness, and soluble solids that were always higher in INT than Control like the year 2017. Dry residue, pH, and titratable acidity were not influenced by fertilization and mulch. The interaction Fertilization × Mulch was not significant for any of the variables. It is worth pointing out that the higher values of color components, firmness, and TSS were detected always in INT X BDM and the lowest ones in Control × Bare (standard practice of farm).

The effect of treatments on antioxidant compounds and antioxidant activity assessed in tomato cv Kendras, in 2018 is reported in Table 9. Fertilization effect was significant only on flavonoids content and antioxidant activity that were higher in INT than Control. The other antioxidant compounds, ascorbic acid, polyphenols, and lycopene were not influenced by the fertilization. Soil mulching effect was significant only on ascorbic acid content, which was higher in bare soil than mulched soil. The interaction Fertilization × Mulch was not significant for any of the variables.

Table 9. Antioxidant compounds and antioxidant activity of tomato fruits cv Kendras, in 2018, as influenced by the mean effects of fertilization, mulch, and their interaction.

Treatments	Ascorbic Acid (mg 100 g^{-1} f.w.)	Polyphenols (mg GAE 100 g^{-1} f.w.)	Flavonoids (mg CE 100 g^{-1} f.w.)	Antioxidant Activity (mg TE 100 g^{-1} f.w.)	Lycopene (mg kg^{-1} f.w.)
Fertilization					
INT	22.9	44.8	5.47 a	183.9 a	44.8
Control	21.4	43.7	4.59 b	159.2 b	43.8
Mulch					
BDM	21.2 b	42.5	4.86	168.6	44.7
Bare	23.1 a	46.0	5.20	174.5	43.9
Significance					
Fertilization	n.s.	n.s.	*	*	n.s.
Mulch	*	n.s.	n.s.	n.s.	n.s.
Fertiliz. × Mulch	n.s.	n.s.	n.s.	n.s.	n.s.

n.s.: not significant differences; * significant differences to Fisher's Test at $p < 0.05$; different letters within each column indicate significant differences according to Tukey HSD Test ($p = 0.05$). INT = integration of solid digestate and mineral fertilizers; Control = farm mineral fertilization; BDM = biodegradable mulch; Bare = bare soil.

4. Discussion

During the vegetative phase, dry matter, as well as nutrients, were allocated mainly in the leaves and stems, which together constituted the largest portion of the total plant dry weight [2,4,33]. The peak of dry matter accumulation in leaves and stems was reached 12 weeks after the transplanting in both the two cultivars tested separately in the two years. From week 12 onward, due to the increase of fruit setting and growth, fruits became the major sinks of dry weight and nutrients that were remobilized from leaves and stems compartments to be reallocated to the fruits until to the harvest [33].

Partitioning of dry biomass between fruits and aboveground vegetative structures at harvest varied in the two years. In 2017, dry biomass in fruits ranged from 68.3% in INT × Bare, to 73% in INT × BDM and Control × BDM to 77.3% in Control × Bare. Hartz and Bottoms [4] found a partition ranging between 60 and 64% in different fields in California. Instead, in 2018, dry biomass accumulated in fruits decreased in the range 50%–56.5% not attributable to specific attitude of cv Kendras. In 2018, dry weight in fruits decreased at the harvest time below the level detected in the preceding phase. This abnormal trend is also more evident, taking into account that dry weight in fruits of Kendras at week 12 was about 300 g per plant, a content higher than that measured in cv Heinz 5508 in 2017. Our explanation is that a mistake was made failing to determine the right harvest time by postponing it. Indeed, we can exclude destructive weather events that were not recorded in the period between weeks 12 and 15, as well as no pest damage occurring. Therefore, at the moment of the harvest the first 1–2 trusses of fruits were over-ripen and rotten on soil determining a loss of marketable yields.

In both the years, the two fertilization strategies determined yields not significantly different. In INT treatment, the total amount of N was slightly reduced from 360 to 300 kg ha^{-1} and half of this amount was supplied as solid digestate; besides, P fertilization was cut of 135 kg ha^{-1}. Many authors agree that the nitrogen needs of tomato to guarantee yields above 100–130 Mg ha^{-1}, are 200 kg N ha^{-1} [2–4,34]. Therefore, it's plausible to further lower the N amount supplied in the hosting farm by cutting the quote of N chemical fertilizers while supplying the most part of N through the solid digestate. As reported by different papers [6,7,35] it is possible a continuous use of digestate in vegetable crop successions. In particular, crops cultivated in spring-summer cycles can be fertilized only with digestate without the integration of chemical fertilizers.

The mulch effect influenced significantly the yields in 2017. The temperature and humidity modification of soil contribute to improve microbial activities and plant nutrition with positive consequences on yields [11,36,37]. As technical information, we signal that the biodegradable film did not hamper the mechanical harvesting during which its fragments were dispersed on soil. The successive tillage incorporated biodegradable fragments into soil, where they underwent complete mineralization.

Morra et al. [38] focused the economic impact of the innovations tested by analyzing the cropping costs during the trials carried out in 2017 and 2018. Farmer's gross income improved, compared to the standard farm practice, when chemical fertilizers were reduced, biodegradable mulch was adopted, and tomato yields overcame 120 Mg ha^{-1}.

In addition to the positive environmental and economic effects, the innovations tested generally led to an improvement in quality of fruits. The results showed that INT and BDM treatments improved the industrial quality of the fruit. As far as color is concerned, since the "L" component is linked to brightness, "a" (redness) to red if positive and green if negative, while "b" (yellow) is linked to yellow if positive and blue if negative, the positive and higher values of the color components indicate that INT fertilization with BDM mulching produced more intense orange-red fruits than Control in bare soil. Likewise, an increase in firmness and, therefore, a better consistency of the fruit was also observed. Total soluble solids and titratable acidity are parameters that influence the organoleptic quality of the tomato such as sweetness and acidity. The results showed an increase of quality also in terms of TSS and TA, the latter only for 2017. Moreover, for the trial carried out in 2017, fruits from plant grown under the combination of fertilization with digestate and soil mulching exhibited the highest values for quality in terms of firmness and titratable acidity. It is worth noting that the results from 2017 and 2018 trials indicate that both INT fertilization and BDM or their combination produced positive effects or did not lead to detrimental changes in the fruit quality for the considered parameters.

As far as the fertilization strategy tested, the literature review conducted for this article did not reveal similar previous studies. With regard to the mulch effect, the results obtained are in agreement with what reported by our previous studies on tomatoes carried out under tunnel or in greenhouse in South Italy on Coronel F1 and Kero F1 tomato cultivars [20,39].

Tomatoes has been identified as a functional and nutraceutical food because it is an excellent source of bioactive antioxidant compounds, namely phenolics, vitamin C, and lycopene [40]. These compounds have been associated with several health benefits [41] comprising the ability to protect the body against cancer [42,43], to reduce inflammation [44], and to support the heart health by lowering total cholesterol, LDL cholesterol, and triglycerides, and the risk of atherosclerosis [45].

The experimental factors tested, fertilization and mulching, influenced the antioxidant content and antioxidant activity of tomato fruits in the 2017 differently than in the 2018 crop cycle.

In 2017, the ascorbic acid content did not change depending on the type of fertilization or the presence of mulch, and the lowest content was found in fruits from plants grown under the combination Control × Bare treatment; instead, in 2018, it showed a slight decrease in mulched soil compared to bare soil. As reported by many authors [46,47] solar radiation can directly influence the ascorbic acid content and changes in water availability are an important factor [48].

In 2017, total phenolic content increased both under INT fertilization and BDM treatments and the lowest content was found in fruits from plant grown under the combination Control × Bare, while in 2018, it was not affected by the different treatments or their interaction.

As far as flavonoid content is concerned, INT fertilization applied in the same plots in the two years, produced a slight increase in 2018 with respect to 2017. This could be ascribed to the different varieties used. However, according to Mitchell et al. [49], in an investigation of ten years in organic vs. conventional management systems, the level of flavonoid in tomato fruits increased over time in samples from the organic crop system, whereas the levels of flavonoids did not vary significantly in the conventional management. It is possible our data started to reflect the influence of repeated supply of organic carbon by digestate.

Antioxidant activity is correlated with the total polyphenol content; the flavonoids are a class of polyphenols. It can be speculated that the increase in antioxidant activity could alternatively have depended on these different classes of polyphenols in 2017 and 2018.

In both years, the lycopene content of tomato fruits did not change depending on fertilization, while it increased in fruits from plants grown in the presence of the biodegradable mulching only for 2017.

With reference to the effect of Mater-Bi mulch on lycopene, polyphenols, and antioxidant activity, similar results were obtained in our previous studies carried out under tunnel or in greenhouse in South Italy on Coronel F1 and Kero F1 tomato cultivars [20,39]. In this research, it was found that the fruits from plots with Mater-Bi biodegradable black films showed higher concentration of both the antioxidants analyzed and the antioxidant activity in comparison to LDPE and not mulched soil.

According to Hart and Scott [50] the content of tomato antioxidants depends on a lot of factors, especially on cultivars, stage of maturity and growing conditions. Valsikova et al. [51] reported a significant effect of the year, mulching film and variety on the content of ascorbic acids, polyphenols and antioxidant capacity. These authors stated that the quantity and quality of these phytochemicals as well as yields of tomato fruits depend greatly on environmental conditions, agronomic interventions, and genotype.

5. Conclusions

The results above discussed can be conclusively resumed as follows: (a) it is possible to reduce input of chemical N-P fertilizers, replacing them with solid digestate; on average, yields were the same of standard mineral fertilization. Based on this first evidence, the core of the N supply to tomato could be constituted by solid digestate, while the use of N fertilizers could only integrate N via fertigation and up to a total amount around 200 kg ha^{-1}. Similarly, the P supply should be furtherly reduced to 100 kg ha^{-1} in order to avoid the useless amount of phosphatic fertilizers added until now. These savings of fertilizers could be implemented in the farm practice with beneficial effects in costs/gross income balance; (b) the biodegradable mulch remained intact on the soil enough to leave time for tomato plants to completely expand on soil and compete with weeds. Biodegradable mulch did not hamper the mechanical harvesting; the film was easily ripped and dispersed on the soil where it was successively buried by plowing without costs for its collection and disposal. Mulch improved plant production in 2017 but not significantly in 2018. When digestate was used, soil mulching favored a release of nutrients from the organic fertilizer able to sustain plant development as well as mineral fertilization carried out in the farm. Finally, we can confirm our preceding results regarding the effect of the biodegradable Mater-Bi based mulch on the quality of fruits. In both the cultivars, we detected a significant improvement of fruit quality parameters interesting the processing industry, namely, fruit color and firmness, total soluble solids, titratable acidity. The fertilization with the digestate and reduced rates of NP fertilizers particularly favored these effects of mulch.

Author Contributions: Conceptualization, L.M. and E.C.; methodology, L.M., A.S., F.M., L.d.P.; software, L.M. and L.d.P.; investigation, E.C., M.B., S.B., A.S., F.M.; writing—original draft preparation, L.M. and L.d.P.; writing—review and editing, L.M., L.d.P.; funding acquisition, L.M. All authors have read and agreed to the published version of the manuscript.

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References

1. ISTAT Agricoltura. 2020. Available online: https://www.istat.it/it/agricoltura?dati (accessed on 28 May 2020).
2. Tei, F.; Benincasa, P.; Guiducci, M. Critical nitrogen concentration in processing tomato. *Eur. J. Agron.* **2002**, *18*, 45–55. [CrossRef]
3. Elia, A.; Conversa, G. Agronomic and physiological responses of a tomato crop to nitrogen input. *Eur. J. Agron.* **2002**, *40*, 64–74. [CrossRef]
4. Hartz, T.K.; Bottoms, T.J. Nitrogen requirements of drip irrigated processing tomato. *HortScience* **2009**, *44*, 1988–1993. [CrossRef]
5. Action Plan of Campania Region For zones Vulnerable to Nitrate Pollution from Agricultural Sources. 2007. Available online: http://www.sito.regione.campania.it/burc/pdf07/burc16or_07/del209_07.pdf (accessed on 30 May 2020).
6. Maucieri, C.; Nicoletto, C.; Caruso, C.; Sambo, P.; Borin, M. Effects of digestate solid fraction fertilization on yield and soil carbon dioxide emission in a horticulture succession. *Ital. J. Agron.* **2017**, *12*, 1116–1123.
7. Nicoletto, C.; Dalla Costa, L.; Sambo, P.; Zanin, G. Distillery anaerobic digestion residues as fertilizers for field vegetable crops: Performance and efficiency in md-term successions. *Agronomy* **2019**, *9*, 463. [CrossRef]
8. Ronga, D.; Caradonia, F.; Parisi, M.; Bezzi, G.; Allesina, G.; Pedrazzi, S.; Francia, E. Using digestate and biochar as fertilizers to improve processing tomato production sustainability. *Agronomy* **2020**, *10*, 138. [CrossRef]
9. Kasirajan, S.; Ngouajio, M. Polyethylene and biodegradable mulches for agricultural applications: A review. *Agron. Sustain. Dev.* **2012**, *32*, 501–529. [CrossRef]
10. Steinmetz, Z.; Wollmann, C.; Schaefer, M.; Buchmann, C.; David, J.; Tröger, J.; Muñoz, K.; Frör, O.; Schaumann, G.E. Plastic mulching in agriculture. Trading short-term agronomic benefits for long term soil degradation? *Sci. Total Environ.* **2016**, *550*, 690–705. [CrossRef]
11. Martin-Closas, L.; Costa, J.; Pelacho, A.M. Agronomic effects of biodegradable films on crop and field environment. In *Soil Degradable Bioplastics for a Sustainable Modern Agriculture*; Malinconico, M., Ed.; Springer-Verlag GmbH: Berlin/Heidelberg, Germany, 2017; pp. 67–107.
12. Li, C.; Moore-Kucera, J.; Lee, J.; Corbin, A.; Brodhagen, M.; Miles, C.; Inglis, D. Effects of biodegradable mulch on soil quality. *Appl. Soil Ecol.* **2014**, *79*, 59–69. [CrossRef]
13. Sintim, H.Y.; Bandopadhyay, S.; English, M.E.; Bary, A.I.; DeBruyn, J.M.; Schaeffer, S.M.; Miles, C.A.; Reganold, J.P.; Flury, M. Impacts of biodegradable plastic mulches on soil health. *Agric. Ecosyst. Environ.* **2019**, *273*, 36–49. [CrossRef]
14. Sintim, H.Y.; Bary, A.I.; Hayes, D.G.; Wadsworth, L.C.; Anunciado, M.B.; English, M.E.; Bandopadhyay, S.; Schaeffer, S.M.; DeBruyn, J.M.; Miles, C.A.; et al. In situ degradation of biodegradable plastic mulch films in compost and agricultural soils. *Sci. Total Environ.* **2020**, *727*, 138668. [CrossRef] [PubMed]
15. Ghimire, S.; Flury, M.; Scheenstra, E.J.; Miles, C.A. Sampling and degradation of biodegradable plastic and paper mulches in field after tillage incorporation. *Sci. Total Environ.* **2020**, *703*, 135577. [CrossRef] [PubMed]
16. Macua, J.I.; Jiménez, E.; Suso, M.L.; Gervas, C.; Lahoz, I. The future of processing tomato crops in the Ebro valley lies with the use of biodegradable mulching. *Acta Hortic.* **2013**, *971*, 143–146. [CrossRef]
17. Martín-Closas, L.; Bach, M.A.; Pelacho, A.M. Biodegradable mulching in an organic tomato production system. *Acta Hortic.* **2008**, *767*, 267–274. [CrossRef]
18. Moreno, C.; Mancebo, I.; Tarquis, A.M.; Moreno, M.M. Univariate and multivariate analysison processing tomato quality under differentmulches. *Sci. Agric.* **2014**, *71*, 114–119. [CrossRef]
19. Cozzolino, E.; Morra, L.; Petriccione, M. Influenza della pacciamatura sulla qualità del pomodoro da trasformazione. In Proceedings of the XII Meeting AISSA, Sassari, Italy, 6–7 November 2014; University of Sassari: Sassari, Italy, 2014; pp. 21–23.
20. Sekara, A.; Pkluda, R.; Cozzolino, E.; del Piano, L.; Cuciniello, A.; Caruso, G. Plant growth, yield, and fruit quality of tomato affected by biodegradable and non degradable mulches. *Hort. Sci.* **2019**, *46*, 138–145. [CrossRef]

21. Morra, L.; Cozzolino, E.; Cerrato, D.; Bilotto, M.; Mignoli, E.; Coppola, R.; Leone, V.; Petriccione, M.; Pasquariello, M.S.; Parillo, R.; et al. Rese produttive e qualità dei frutti di melone retato (*Cucumis melo* L) allevato sotto tunnel in prove del 2014 e 2015. In *Risultati del Progetto di Sostituzione Delle Pacciamature in Polietilene Con Quelle Biodegradabili in Mater-Bi per Colture Orticole e Frutticole Sotto Serra: Valutazioni Agronomiche ed Economiche*; Morra, L., Cerrato, D., Cozzolino, E., Eds.; Editore ADV Sinopia Scarl: Caserta, Italy, 2015; pp. 40–56.
22. Morra, L.; Bilotto, M.; Cerrato, D.; Coppola, R.; Leone, V.; Mignoli, E.; Pasquariello, M.S.; Petriccione, M.; Cozzolino, E. The Mater-Bi biodegradable film for strawberry (Fragaria x ananassa Duch.) mulching: Effects on fruityeld and quality. *Ital. J. Agron.* **2016**, *731*, 203–206.
23. AOAC International. *Official Methods of Analysis of AOAC International. AOAC International*; Secs. 942.15; AOAC International: Washington, DC, USA, 1995.
24. AOAC International. *Official Methods of Analysis of AOAC International. AOAC International*; Secs. 967.21; AOAC International: Washington, DC, USA, 2002.
25. Jagota, S.K.; Dani, H.M. A new Colorimetric Technique for the estimation of vitamin C using Folin Phenol Reagent. *Anal. Biochem.* **1982**, *127*, 178–182. [CrossRef]
26. Periago, M.J.; Rincon, F.; Aguera, M.D.; Ros, G. Mixture approach for optimizing lycopene extraction from tomato and tomato products. *J. Agric. Food Chem.* **2004**, *52*, 5796–5802. [CrossRef]
27. Kaur, C.; Walia, S.; Nagal, S.; Walia, S.; Singh, J.; Singh, B.B.; Saha, S.; Dingh, B.; Kalia, P.; Jaggi, S.; et al. Functional quality and antioxidant composition of selected tomato (*Solanum lycopersicon* L) cultivars grown in Northern India. *LWT Food Sci. Technol.* **2013**, *50*, 139–145. [CrossRef]
28. Singleton, V.L.; Rossi, J.A. Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. *Am. J. Enol. Vitic.* **1965**, *16*, 144–158.
29. Zhishen, J.; Mengcheng, T.; Jianming, W. The determination of flavonoid contents in mulberry and their scavenging effects on superoxide radicals. *Food Chem.* **1999**, *64*, 555–559. [CrossRef]
30. Pekal, A.; Pyrzynska, K. Evaluation of aluminium complexation for Flavonoid content assay. *Food Anal. Methods* **2014**, *7*, 1776–1782. [CrossRef]
31. Brand-Williams, W.; Cuvelier, M.E.; Berset, C. Use of a free radical method to evaluate antioxidant activity. *LWT Food Sci. Technol.* **1995**, *28*, 25–30. [CrossRef]
32. Ozcelik, J.H.; Lee, J.H.; Min, D.B. Effects of light, oxygen and pH on the adsorbance of 2,2-Diphenyl-1-picrilhydrazyl. *J. Food Sci.* **2003**, *68*, 487–490. [CrossRef]
33. Ozores-Hampton, M.; Di Gioia, F.; Sato, S.; Simonne, E.; Morgan, K. Effects of nitrogen rates on nitrogen, phosphorus, and potassium partitioning, accumulation and use efficiency in seepage-irrigated fresh market tomatoes. *HortScience* **2015**, *50*, 1636–1643. [CrossRef]
34. Ronga, D.; Parisi, M.; Pentangelo, A.; Mori, M.; Di Mola, I. Effects of nitrogen management on biomass production and dry matter distribution of processing tomato cropped in Southern Italy. *Agronomy* **2019**, *9*, 855. [CrossRef]
35. Alburquerque, J.A.; De la Fuente, C.; Campoy, M.; Carrasco, L.; Najera, I.; Baixauli, C.; Caravaca, F.; Rolda, A.; Cegarra, J.; Bernal, M.P. Agricultural use of digestate for horticultural crop production and improvement of soil properties. *Eur. J. Agric.* **2012**, *43*, 119–128. [CrossRef]
36. Cirujeda, A.; Aibar, J.; Anzalone, A.; Martin-Closas, L.; Meco, R.; Moreno, M.M.; Pardo, A.; Pelacho, A.M.; Rojo, F.; Royo-Esnal, A.; et al. Biodegradable mulch instead of polyethylene for weed control of processing tomato production. *Agron. Sustain. Dev.* **2012**, *32*, 889–897. [CrossRef]
37. Moreno, M.M.; Cirujeda, A.; Aibar, J.; Moreno, C. Soil thermal and productive repsonses of biodegradable mulch materials in a processing tomato (*Lycopersicon esculentum* Mill.) crop. *Soil Res.* **2016**, *54*, 207–215. [CrossRef]
38. Morra, L.; Fagnano, M.; Cozzolino, E.; Bilotto, M.; Fiorentino, N.; Pergamo, R. Pacciamatura e nutrizione: Innovazioni per il pomodoro. *L'Inf. Agric.* **2019**, *21*, 48–52.
39. Cozzolino, E.; Sekara, A.; Pokluda, R.; del Piano, L.; Cuciniello, A.; Caruso, G. Plant growth, yield, fruit quality and residual biomass composition of tomato as affected by mulch type. *Acta Hortic.* **2020**, *1271*, 465–472. [CrossRef]
40. Raffo, A.; Leonardi, C.; Fogliano, V.; Ambrosino, P.; Salucci, M.; Gennaro, L.; Bugianesi, R.; Giuffrida, F.; Quaglia, G. Nutritional value of cherry tomatoes (*Lycopersicon esculentum* cv Naomi F1) harvested at different ripening stages. *J. Agric. Food Chem.* **2002**, *50*, 6550–6556. [CrossRef]
41. Canene-Adams, K.; Campbell, J.K.; Zaripheh, S.; Jeffery, E.H.; Erdman, J.W. The tomato as a functional food. *J. Nutr.* **2005**, *135*, 1226–1230. [CrossRef] [PubMed]
42. Campbell, J.K.; Canene-Adams, K.; Lindshield, B.L.; Boileau, T.W.M.; Clinton, S.K.; Erdman, J.W. Tomato phytochemicals and prostate cancerrisk. *J. Nutr.* **2004**, *134*, 3486–3492. [CrossRef]
43. Bhuvaneswari, V.; Nagini, S. Lycopene: A review of its potential as an anticancer agent. *Curr. Med. Chem. Anti-Cancer Agent* **2005**, *5*, 627–635. [CrossRef]
44. Palozza, P.; Parrone, N.; Catalano, A.; Simone, R. Tomato lycopene and inflammatory cascade: Basic interactions and clinical implications. *Curr. Med. Chem.* **2010**, *17*, 2547–2563. [CrossRef]
45. Ried, K.; Falker, P. Protective effect of lycopene on serum cholesterol and blood pressure: Meta-analyses of intervention trials. *Maturitas* **2011**, *68*, 299–310. [CrossRef]

46. Dumas, Y.; Dadomo, M.; Di Lucca, G.; Grolier, P. Effects of environmental factor and agricultural tecniques on antioxidant content of tomatoes. *J. Sci. Food Agric.* **2003**, *83*, 369–382. [CrossRef]
47. Gautier, H.; Massot, C.; Stevens, R.; Serino, S.; Genard, M. Regulation of tomato fruit ascorbate content is more highly dependent of fruit irradiance than leaf irradiance. *Ann. Bot.* **2009**, *103*, 495–504. [CrossRef]
48. Raffo, A.; La Malfa, G.; Fogliano, V.; Maiani, G.; Quaglia, G. Seasonal variation in antioxidant component of cherry tomatoes (*Lycopersicon esculentum* cv Naomi F1). *J. Food Compos. Anal.* **2006**, *19*, 11–19. [CrossRef]
49. Mitchell, A.E.; Hong, Y.; Koh, E.; Barret, D.M.; Bryant, D.E.; Denison, R.F.; Kaffa, S. Ten-year comparison of the influenced of organic and conventional crop management practices on the content of flavonoids in tomatoes. *J. Agric. Food Chem.* **2007**, *55*, 6154–6159. [CrossRef] [PubMed]
50. Hart, D.J.; Scott, K.J. Development and evaluation of HPLC method for the analysis of carotenoids in foods and the measurement of the carotenoids content of vegetables and fruit commonly consumed in the UK. *Food Chem.* **1995**, *54*, 101–111. [CrossRef]
51. Valsikovà, M.; Mlcek, L.; Snopek, L.; Rehus, M.; Skrovankova, S.; Jurikova, T.; Sumkzynsky, D.; Paulen, O. Monitoring of bioactive compounds of tomato cultivars as affected by mulching film. *Sci. Agric. Bohem.* **2018**, *49*, 267–273. [CrossRef]