

## Article

# Effect of Organic and Mineral Soil Additives on Asparagus Growth and Productivity in Replant Soils

Roxana Djalali Farahani-Kofoet <sup>1,\*</sup> , Franziska Häfner <sup>2</sup> and Carmen Feller <sup>1</sup><sup>1</sup> Leibniz Institute of Vegetable and Ornamental Crops (IGZ), 14979 Großbeeren, Germany; feller@igzev.de<sup>2</sup> Soil Quality and Soil Use Research Group, Department of Agroecology and Environment, Agroscope, 8046 Zürich, Switzerland; franziska.haefner@agroscope.admin.ch

\* Correspondence: kofoetr@igzev.de

**Abstract:** The repeated cultivation of asparagus in the same field can severely reduce yield. A complex of predominantly microbial causes is suspected. Limited plant development, establishment problems, and yield loss may occur, particularly in light sandy soils. In order to address this replant problem and evaluate alternative cultivation conditions, two asparagus fields were treated with different supplements and were cultivated for 5 years to investigate their impact on yield. The results from the pot trials using soils from these fields are presented, along with the field trial findings. The trials included the incorporation of mushroom substrate (champost), Fimonit (clay mineral), mustard meal (biofumigation), and Micosat F Uno (including arbuscular mycorrhizal fungi, *Trichoderma viride*, and rhizosphere bacteria species). In the pot trials, the sterilised soil exhibited a growth benefit over the original soil. However, the tested additives had no significant effects in the short period of 8 weeks. At one of the tested field sites, the marketable asparagus yields following champost, Fimonit, biofumigation, and Micosat treatments were 14, 6, 16 and 12% higher than that of the control soil, respectively, but no significant differences in treatment effect were observed in the second test field. Biofumigation using mustard meal and champost was most successful in reducing the impact of replanting on yields.

**Keywords:** champost; clay minerals; biofumigation; arbuscular mycorrhizae; asparagus pot trial; asparagus field trial



**Citation:** Djalali Farahani-Kofoet, R.; Häfner, F.; Feller, C. Effect of Organic and Mineral Soil Additives on Asparagus Growth and Productivity in Replant Soils. *Agronomy* **2023**, *13*, 1464. <https://doi.org/10.3390/agronomy13061464>

Academic Editor: Claudio Ciavatta

Received: 3 May 2023

Revised: 17 May 2023

Accepted: 23 May 2023

Published: 25 May 2023



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## 1. Introduction

The fleshy green, violet, and white shoots of *Asparagus officinalis* L. are a highly nutritious and low-calorie comestible which is popular with consumers worldwide. Asparagus, unlike most vegetables, is a perennial crop that can be cultivated at the same site for approximately 12 years and which grows well in well-drained soil in subtropical and temperate climates [1]. Universally, after 6 to 7 years of production, a gradual decrease in yield is observed, known as the asparagus decline. This makes the maintenance of the stand no longer profitable, leaving farmers with the option of having to cultivate new land that is most likely not suitable for asparagus production or replanting in abandoned asparagus fields. When asparagus is replanted in fields previously used for this crop, this is often associated with significant reductions in yield rates [2,3]. Economically relevant drops in marketable yields of up to 30% [4] or even 60% [5] have been reported. Numerous studies and reviews dealing with asparagus decline and the replant problem highlight the issues of yield and productivity in asparagus farms. The problem is attributed to the increase in soil-borne pathogens, specifically *Fusarium* spp. and *Phytophthora* rot, as well as the production of autotoxins and allelochemicals from the root exudates of old asparagus plants. Many studies looking to address the replant problem have investigated types of biotic and/or abiotic stress [1,3,6–14]. Noperi-Mosqueda et al. [15] studied the effect of asparagus decline on leaf nutrients accumulation and on the yield and quality of

the spears and found significant changes in macronutrient concentrations, but B deficiency and Fe toxicity, as well as higher phenolic compounds in replant soils, could contribute to decline syndrome. Soil sterilisation has been demonstrated to reduce post-replanting yield decline [16]. Therefore, a predominantly microbial-cause complex with an imbalance in bacterial relative abundances and an accumulation of root-damaging soil micro-organisms has been speculated [17–19]. Furthermore, a positive correlation has been reported between yield decline and light sandy soils [20].

Asparagus farmers in Germany depend on old asparagus fields due to a lack of arable land. Chemical soil treatment—not approved in Germany until 2019—could be effective, but few studies have investigated this approach to the best of our knowledge. Since crop rotation and land swapping are not possible, improving soil properties by supplementation could be a key solution. Biotic additives can be used as potential biocontrol agents in a sustainable and environmentally friendly asparagus cultivation strategy. The addition of organic matter (e.g., compost) or the use of pathogen-suppressing micro-organisms can also positively impact soil health [2]. Blok et al. [21] achieved a significant decrease in soil infestation levels regarding *Fusarium* pathogens when incorporating grass into moist soil and covering it with airtight plastic. Borrego-Benjumea et al. [22] incorporated pelleted poultry manure into soils and achieved a disease severity reduction of ca. 50% in greenhouse experiments. Inoculation with beneficial micro-organisms (e.g., *Pseudomonas*, *Streptomyces*, *Trichoderma*, arbuscular mycorrhizae) has been shown to promote growth in old asparagus fields [9,23]. Matsubara et al. [24] found plant growth promotion and the suppression of *Fusarium* crown rot after inoculating asparagus plants with arbuscular mycorrhizal fungus. The use of mustard meal, known as biofumigation, has also been shown to suppress soil-borne pathogens. The isothiocyanates produced during the degradation of the plant parts of the *Brassicaceae* family (mainly mustard flour) have antimicrobial, fungicidal, and nematocidal properties [25]. Lopez-Moreno et al. [26] compared the effect of biofumigation with *Brassica* pellets, biofumigation with chicken manure pellets, and the disinfection of the soil using Dazomet and concluded that the *Brassica* pellets and the chemical, Dazomet, were the most effective against the damage caused by the decline syndrome.

In order to solve the asparagus replant problem, we set up two long-term field trials in East Germany, in which we investigated whether the addition of compost (mushroom compost), mustard meal, arbuscular mycorrhizae, or beneficial micro-organisms (to the soil) affected the productivity of asparagus. Greenhouse pot experiments were performed previously. We hypothesized that the soils from the selected field trials would have a decline effect on the asparagus plants and the yield, and the application of additives would overcome the replant problem.

## 2. Materials and Methods

### 2.1. Site Selection, History, and Soil Properties

Two sites, A and B, were selected for pot and long-term field trials. Site A, at the research station of the Leibniz Institute of Vegetable and Ornamental Crops in Grossbeeren, Germany (52°33' N, 13°22' E), had a five-year cultivation break with wheat before asparagus replantation. Site B, an asparagus farm in Beelitz, Germany (52°17' N, 12°49' E), was cultivated with wheat for one year in between asparagus cultivation and replanting. From each site, 16 samples from the top soil layer (0–30 cm) were taken with a soil probe (18 mm in diameter) and pooled with thorough mixing. The presence and abundance of pathogenic fungi were analysed externally (Scientia Terrae Research Institute, Antwerp, Belgium) using the DNA Multiscan technique, which uses an array of genus- and species-specific DNA fragments to detect and quantify a variety of mainly plant-related pathogenic fungi. The intensity of the DNA hybridisation signal is scored from 0 (not detected) to 6 (strongest signal). Furthermore, the basic properties of the soils were determined in a service laboratory (Table 1). *Fusarium solani* and *Pythium ultimum* were detected in the soils of both sites, while *F. oxysporum* and *P. irregulare* were only detected in site B, with strong

signals observed for site B and moderate for site A. Therefore, the presence of pathogens was determined, and the soils were considered suitable for further replanting experiments.

**Table 1.** Properties of the soil samples from sites A and B before the experiment (2018), including the analysis of pathogenic organisms.

Site	Clay (%)	Soil Class	OS (%)	pH	DNA Multiscan Hybridisation Signal					
					<i>Fusarium</i> sp.	<i>F. oxysporum</i>	<i>F. solani</i>	<i>Pythium</i> sp.	<i>P. ultimum</i>	<i>P. irregulare</i>
A	7	Sand	1.2	6.1	6	0	4	4	4	0
B	3.8	Sand	1.8	5.7	6	4	6	6	6	4

OS, organic substance.

## 2.2. Testing Supplemented Soils from Sites A and B through Pot Trials

To assess the impact of mineral and organic additives on the development of young ‘Gijnlim’ (Limgroup B.V., Horst, The Netherlands), ‘Ravel’, and ‘Ramires’ (Südwestdeutsche Saatzucht GmbH & Co. KG, Rastatt, Germany) asparagus plants, pot trials with soil from sites A (soil A) and B (soil B) were carried out (Table 2). Overall, 100 seeds per cultivar were placed in soil-filled trays (40 × 40 × 40 mm) (Fruhstorfer Erde/Type P soil; HAWITA Gruppe GmbH, Vechta, Germany) and were left to germinate in a greenhouse for 8 weeks at 20/16 °C for the day and night temperatures. Seedlings at BBCH stage 2–3 were planted in 2 L pots (2100 g soil) with each of the treatments described below. The plants were watered and fertilised with equal amounts of nutrient solution.

**Table 2.** Overview of some important data obtained from the pot trials.

	1. Pot Trial	2. Pot Trial	3. Pot Trial
Treatments	untreated soil, sterilised soil, untreated soil + Fimonit®	untreated soil, sterilised soil, untreated soil + Promot, untreated soil + champost, untreated soil + RhizoVital	untreated soil, sterilised soil, untreated soil + champost, untreated soil + biofumigation
Cultivar	‘Gijnlim’, ‘Ravel’, ‘Ramires’	‘Gijnlim’	‘Gijnlim’
Site	A and B	B	B
Replicates	4	5	6
Model	three-factorial	one-factorial	one-factorial
Weeks	12	12	8

In the first pot trial, we investigated the replant effect by comparing the sterilised and untreated soils. Since soils A and B were contaminated with pathogens, they were steam-sterilised in autoclave bags and dried in a compartment drier at 100 °C for 1 h. Untreated soils served as controls. As a third treatment, we examined the impact of adding clay minerals (1.6 g/L Fimonit® SEAL; FIM Friedland Industrial Minerals GmbH, Friedland, Germany) to untreated soils on the asparagus shoot and root growth. ‘Gijnlim’, ‘Ravel’, and ‘Ramires’ seedlings were planted in prepared pots and cultivated for 8 weeks under greenhouse conditions (see above) in a randomised design. After harvesting the asparagus plants, the shoots and roots were dried to a constant weight in a dry cabinet to determine the dry matter (DM) biomass. Four replicates were performed in this three-factorial trial (3 treatments, 2 sites, and 3 cultivars) for each treatment.

In the second trial, we assessed the effect of the organic additives champost (10 g/L; Biopilzhof); Promot®, containing *Trichoderma harzianum* (100 mg/L; Intrachem Bio Deutschland GmbH & Co. KG, Bad Camberg, Germany), and RhizoVital® 42 TB, containing *Bacillus amyloliquefaciens* strain FZB (200 mg/L; ABiTEP GmbH, Berlin, Germany), on the growth of the ‘Gijnlim’ variant. Since the first pot trial demonstrated greater effects on asparagus growth in soil B, this soil was selected for this pot trial. Untreated, steam-sterilised soil was used as the control. Plants were processed as described above after 8 weeks of growth under greenhouse conditions. Five replicates were performed for each treatment.

The third trial aimed to evaluate the impact of champost (10 g/L) and biofumigation in unsterilised versus steam-sterilised soil B. Untreated soil B served as the control. Uniformly grown 16-week-old ‘Gijnlim’ plants were planted in 2 L pots. For biofumigation, untreated and unsterilised soil B was watered to 60% of its maximum water-holding capacity. Mustard meal (*Brassica juncea*) (3.43 g/L; Kosmalski Herbs & Spices) was then added into the soil and incubated at 23 °C for five days in sealed, airtight PE bags. The application rate was calculated as 0.3% (*wt/wt*), according to [27]. Plants were cultivated from the beginning of June until the end of July in an open-sided greenhouse, being watered uniformly. The number of shoots and length of the longest shoot were recorded weekly. Each treatment was replicated six times.

### 2.3. Testing Supplements in Field Trials

The field trials at sites A and B were set up according to a strip-plot block design (Figure 1) of four replicates per factor combination (soil treatment and asparagus variant). For growth evaluation, measurements were recorded from 60 plants per replicate of factor combination. Each section (factor combination) measured 4 × 9 m and contained two rows of plants. Basic fertiliser (N, 60 kg/ha; P, 37 kg/ha; K, 100 kg/ha; Mg, 35 kg/ha) was administered to the soil of each section, according to asparagus cultivation recommendations. The treatments included (i) control (untreated soil); (ii) champost, 30 t/ha; (iii) Fimonit clay minerals, 5 t/ha; (iv) mustard meal for biofumigation, 5 t/ha; and (v) Micosat F<sup>®</sup> Uno (microgranulate composed of arbuscular mycorrhizae and beneficial root symbiotic micro-organisms; CCS AOSTA Srl), 10 g per plant. All soil treatments were executed on the same day. The champost, Fimonit, and mustard meal were applied uniformly in their respective sections (across both planting rows), and immediately incorporated into the soil (0–40 cm depth) using a deep spade, followed by flattening of the soil surface. The amount of mustard meal to be added was calculated from *in vitro* biofumigation assays [28]. The sections where mustard meal was incorporated into the soil were sealed with transparent film (200- $\mu$ m thickness) for 14 days. Six days after the film was removed, one-year-old ‘Gijnlim’, ‘Ravel’, and ‘Ramires’ asparagus crowns (purchased) were planted in the field section. The Micosat microgranulate treatment was applied directly into the rows during planting. The trial lasted from the spring of 2018 to the summer of 2022. During this period, asparagus cultivation included pest management according to an integrated production approach. In August 2019, the number of green shoots per plant was recorded. In order to produce white asparagus spears, dams were prepared and covered with black film every March (from 2020). When the first shoots surfaced on the ridge, the daily harvest began. Harvested white asparagus shoots were cut at 23 cm and categorised into four classes according to their diameter: >26 mm; 16–26 mm; 12–15.9 mm, and <12 mm. All classes, including shoots with quality issues (rusty, hollow, and crooked shoots), were taken into account for the total yield calculations. For the marketable yield, shoots thinner than 12 mm and those with quality failures were excluded. The duration of harvesting depended on the year. In 2020, 2021, and 2022, it lasted 3, 5, and 6 weeks, respectively, at both sites. Due to a cold spell in the spring of 2019, many of the spears were affected by frostbite, leading to a failed harvest in 2020 for site B. The field trials ended with soil samples being taken from each section of the two sites for DNA Multiscan analysis.

### 2.4. Statistical Analyses

Three-factorial (first pot trial) and one-factorial (second and third trial) pot experiments were evaluated for possible treatment, cultivar, site, and interaction effects using ANOVA in Statistica<sup>®</sup> v13.5 (TIBCO Software Inc., Palo Alto, CA, USA). Significant differences were tested with the Tukey test, with  $p \leq 0.05$  set as the threshold for statistical significance. The data from the two-factorial strip-block field trials were analysed for possible treatment, variety, or interaction effects using the F-test. Mixed models were fitted via the ‘PROC MIXED’ procedure in SAS 9.4 (SAS Institute, Cary, NC, USA). Pairwise significant dif-

ferences between treatments were determined using the Tukey test ( $p \leq 0.05$ ). Variance homogeneity and normal distribution of residuals were previously controlled.

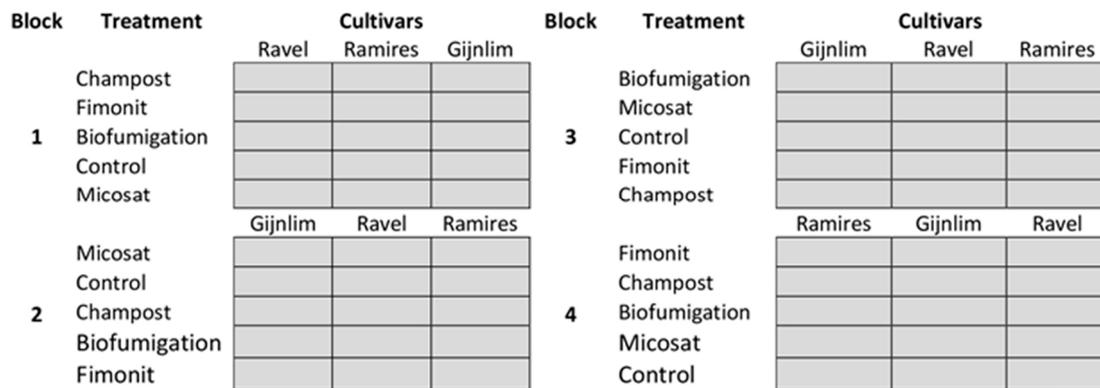


Figure 1. Scheme of the two-factorial strip-plot block design.

### 3. Results

#### 3.1. Pot Trials

In the first pot trial, the steam sterilisation of soils A and B led to a significantly higher root mass when compared to that observed in the untreated or clay-supplemented soil (Table 3). The addition of clay minerals did not result in improved shoot and root growth in the given conditions when compared to the untreated control. The plants appeared to grow better overall in soil A compared with B, regardless of treatment. When comparing the three tested asparagus cultivars across the treatments, no significant variations were observed among them when grown in soil A. However, the results from cultivation in soil B showed that the ‘Gijnlim’ roots developed more than those of the ‘Ramires’ plants.

Table 3. Dry matter (g per plant) from the shoots, roots, and total biomass of the ‘Gijnlim’, ‘Ravel’, and ‘Ramires’ asparagus variants grown in pots with soils A and B 8 weeks after steam sterilisation or clay mineral supplement.

Effect	Shoots (g/Plant)	Roots (g/Plant)	Total Biomass (g/Plant)
Treatment			
Untreated soil	8.3 ± 0.31 a	19.7 ± 0.77 b	27.9 ± 0.95 b
Untreated soil + clay mineral	8.5 ± 0.22 a	19.0 ± 0.75 b	27.4 ± 0.95 b
Steam-sterilised soils	8.1 ± 0.21 a	22.6 ± 0.92 a	30.7 ± 1.02 a
HSD ( $p = 0.05$ )	0.73	1.94	2.01
Source site of soil			
A	8.9 ± 0.16 a	23.1 ± 0.58 a	32.0 ± 0.58 a
B	7.6 ± 0.18 b	17.9 ± 0.55 b	25.5 ± 0.59 b
HSD ( $p = 0.05$ )	0.49	1.31	1.36
Site/cultivar			
A/Gijnlim	8.7 ± 0.21 a	22.8 ± 0.97 a	31.5 ± 0.91 a
A/Ravel	9.1 ± 0.35 a	24.1 ± 1.27 a	33.2 ± 1.28 a
A/Ramires	9.0 ± 0.29 a	22.4 ± 0.82 a	31.3 ± 0.81 a
HSD ( $p = 0.05$ )	1.03	2.74	2.84
B/Gijnlim	8.0 ± 0.29 a	20.0 ± 1.01 a	28.0 ± 1.01 a
B/Ravel	7.5 ± 0.36 a	18.2 ± 0.73 ab	25.7 ± 0.81 ab
B/Ramires	7.4 ± 0.29 a	15.5 ± 0.58 b	22.9 ± 0.67 b
HSD ( $p = 0.05$ )	1.03	2.74	2.84

±Standard error; different letters denote significant differences in each column and the respective effect at  $p \leq 0.05$ .

In the second trial, the roots of the ‘Gijnlim’ plants grown in the steam-sterilised soil B contained significantly more biomass than those of the plants grown with other experimental treatments, including the control (Table 4). No differences were detected between the shoot, root, or total biomass of the plants grown in the soils supplemented with *Trichoderma harzianum* (Promot), *Bacillus amyloliquefaciens* (RhizoVital), champost, or no additive.

**Table 4.** Dry matter (g/plant) from shoots, roots, and total biomass of the ‘Gijnlim’ asparagus variant after 8 weeks of growth in soil B, treated with Promot, champost, RhizoVital, or steam sterilisation.

Soil B Treatment	Shoots (g/Plant)	Roots (g/Plant)	Total Biomass (g/Plant)
Untreated	9.1 ± 0.38 a	18.3 ± 0.57 b	27.3 ± 0.50 b
Untreated + Promot	9.0 ± 0.51 a	18.3 ± 1.55 b	27.2 ± 1.44 b
Untreated + Champost	7.6 ± 0.36 a	19.3 ± 1.83 ab	27.0 ± 2.10 b
Untreated + RhizoVital	8.7 ± 0.49 a	20.5 ± 0.94 ab	29.2 ± 0.79 ab
Steam sterilised	8.9 ± 0.27 a	25.2 ± 1.68 a	34.0 ± 1.74 a
HSD ( $p = 0.05$ )	1.75	5.95	6.13

±Standard error; different letters denote significant differences in each column at  $p \leq 0.05$ .

In the third pot experiment, the biofumigation of the soil resulted in increased shoot and root development in the ‘Gijnlim’ cultivar (Table 5). No differences were noted among the plants grown in the control, champost-supplemented, and steam-sterilised soils in this setting.

**Table 5.** Dry matter (g/plant) from shoots, roots, and total biomass of the ‘Gijnlim’ asparagus variant after 8 weeks of growth in soil B that was either steam sterilised, supplemented with champost, or biofumigated.

Soil B Treatment	Shoots (g/Plant)	Roots (g/Plant)	Total Biomass (g/Plant)
Untreated	2.0 ± 0.09 b	9.1 ± 0.50 b	11.0 ± 0.56 b
Steam sterilised	2.0 ± 0.11 b	10.3 ± 0.33 b	12.3 ± 0.31 b
Untreated + Champost	2.3 ± 0.09 b	10.7 ± 0.67 b	13.0 ± 0.73 b
Untreated + Biofumigation	3.6 ± 0.14 a	16.0 ± 0.60 a	19.6 ± 0.66 a
HSD ( $p = 0.05$ )	0.44	2.15	2.34

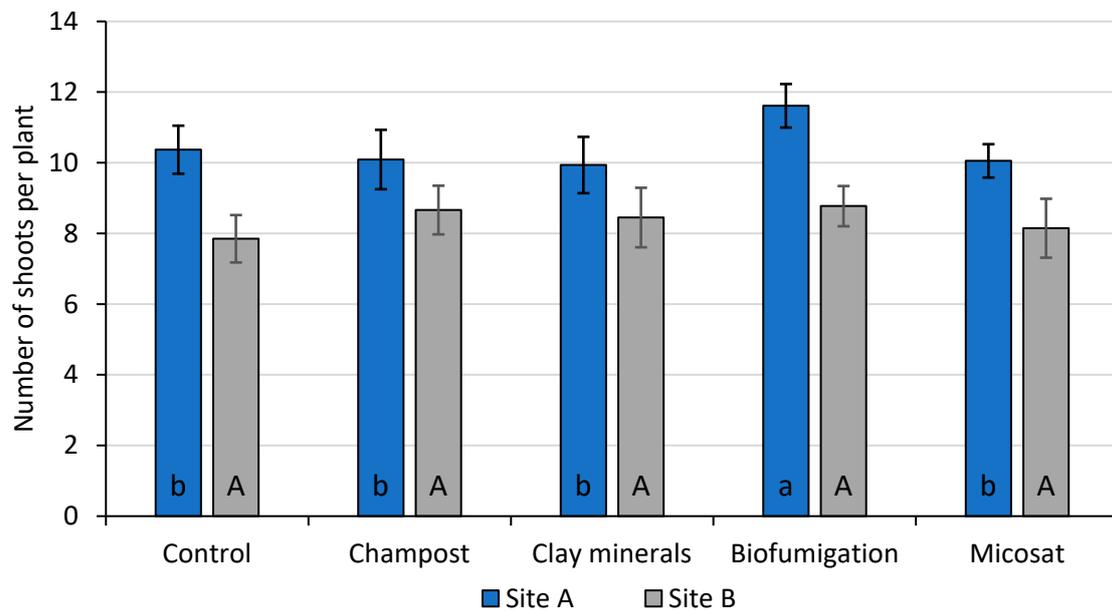
±Standard error; different letters denote significant differences in each column at  $p \leq 0.05$ .

### 3.2. Field Trials

Notable differences in development were already evident by the second year of the field experiment (2019) when the shoots were counted (Figure 2). At site A, the plants grown in the biofumigated soil produced significantly more shoots per plant compared to those grown under other treatment conditions. However, at site B, shoot production did not appear to differ among the plants grown under the different treatments.

The total marketable yield over the 3 years of harvest at site A demonstrated significant treatment and cultivar effects, but no interaction effect was observed between treatment and cultivar (Table 6). Champost treatment and biofumigation resulted in a significantly higher marketable yield for all varieties when compared to the control soil (Figure 3A). The application of clay minerals or Micosat did not appear to affect the yield of the cultivars in the given time. Biofumigation, champost, Micosat, and clay mineral treatment led to 16, 14, 12 and 7% increases in total marketable yield, respectively, when compared to the control conditions (Supplementary Table S1). The impact of biofumigation was significantly greater than that of the clay mineral treatment and the control in the third year of cultivation (2020), and greater than the control in the third and fourth years (2020 and 2021). In the fifth year (2022), the impact of biofumigation had diminished. There was no longer a significant

treatment effect (Figure 3A; Table 6). Across all years of observation, the ‘Gijnlim’ and ‘Ravel’ variants achieved higher yields than ‘Ramires’ (Figure 4A, Supplementary Table S2). In 2022, the year with the longest harvest period, no significant difference was detected among the cultivars ‘Gijnlim’ and ‘Ramires’ (Figure 4A; Table 6).



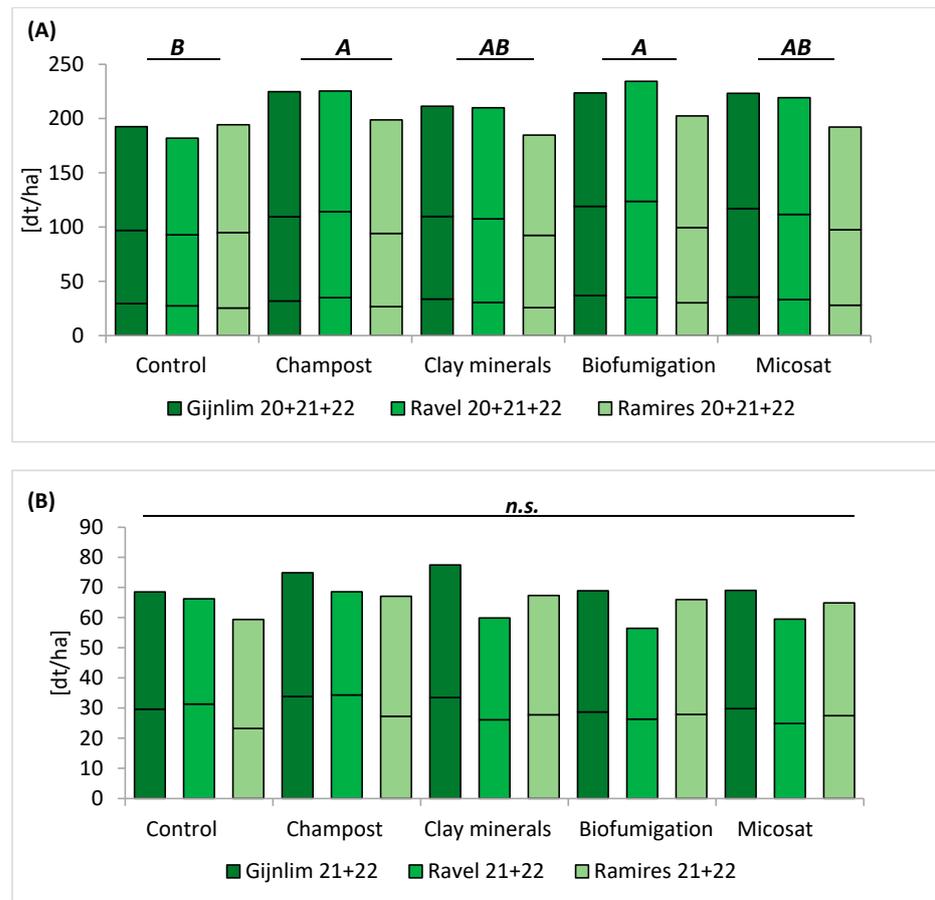
**Figure 2.** Average number of shoots on plants grown in sites A and B in 2019, the second year of asparagus cultivation. The soil treatments included champost, clay minerals, mustard meal (biofumigation), or root treatment with Micosat. Upper- and lower-case letters refer to the respective sites. Different letters denote significant differences between treatments of the same site ( $p \leq 0.05$ ). Error bars represent a 95% confidence interval.

**Table 6.**  $p$ -values for treatment, cultivar, and interaction effects on market yield produced at the two field trial sites, as calculated from the variance analysis.

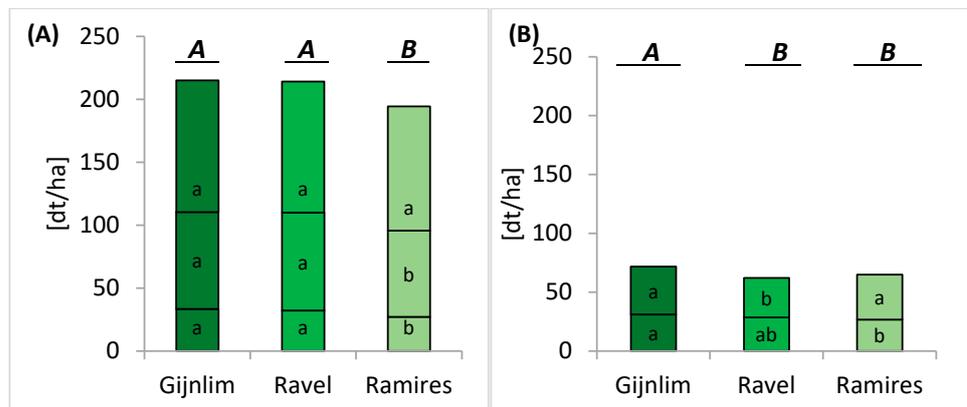
	Site A				Site B		
	2020	2021	2022	Sum 2020–2022	2021	2022	Sum 2021–2022
Treatment	<0.001 *	0.007 *	0.109	0.013 *	0.101	0.486	0.127
Cultivar	<0.001 *	0.001 *	0.388	0.008 *	0.010 *	<0.001 *	<0.001 *
Treatment × cultivar	0.649	0.268	0.877	0.655	0.123	0.815	0.306

\* Significant at  $p \leq 0.05$ .

The marketable yield from site B was lower than that from site A (Figure 3). No statistically significant differences were observed across treatments in site B; however, in soil supplemented with champost and clay minerals, a positive trend was noted for the ‘Gijnlim’ and ‘Ramires’ (Figure 3B) variants. There was a significant cultivar effect across the years (Figure 4B; Table 6): the ‘Gijnlim’ cultivar produced a higher yield than the ‘Ramires’ variant in 2021, whereas, in 2022, the yields of the ‘Gijnlim’ and ‘Ramires’ strains were significantly higher than that of ‘Ravel’ (Figure 4B, Supplementary Table S2). Over both years, the ‘Gijnlim’ variant demonstrated significantly higher yield potential than the other two tested cultivars (Figure 4B).

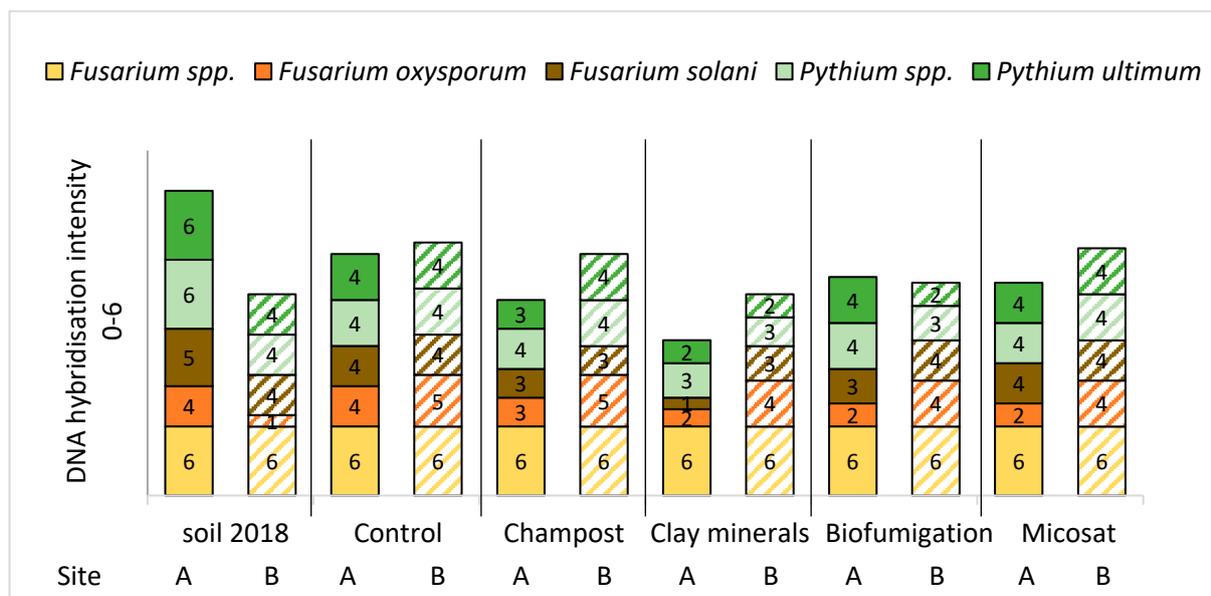


**Figure 3.** The marketable yield of the ‘Gijnlim’, ‘Ravel’, and ‘Ramires’ asparagus cultivars in the field trials for (A) site A and (B) site B. Stacked columns (from bottom to top) indicate the marketable yield of 3 or 2 harvest years, respectively. The soil treatments included champost, clay minerals, mustard meal (biofumigation), or asparagus root treatment with Micosat. Different letters show significant differences in the total sum of marketable yield among the treatments (Tukey test;  $p \leq 0.05$ ). n.s., not significantly different.



**Figure 4.** Marketable yield of the ‘Gijnlim’, ‘Ravel’, and ‘Ramires’ asparagus cultivars in field trials for (A) site A and (B) site B. Stacked columns (from bottom to top) indicate the marketable yield of 3 or 2 harvest years, respectively. Different bold italic upper-case letters indicate significant differences among the cultivars. Different lower-case letters stand for significant differences among the cultivars in each year (Tukey test;  $p \leq 0.05$ ).

After the final harvest, the soils were tested for micro-organisms and *Fusarium* spp., *F. oxysporum*, *F. solani*, *Pythium* spp., and *P. ultimum* were detected at both sites (Figure 5). The lowest signals for these fungi were noted in the soils supplemented with clay minerals. Overall, the samples from site A demonstrated lower signals than those from site B, as observed in the 2018 DNA Multiscan results (Table 2). The pathogen abundance signals were similar in the site B soils supplemented with Micosat and champost. At the same site, the ability of biofumigation to suppress the soil-borne pathogens was comparable to that of clay minerals. The champost treatment of site A soil was more effective than biofumigation. *Fusarium* spp. was detected at the same intensity in all samples (Figure 5).



**Figure 5.** DNA Multiscan hybridisation signal intensities for *Fusarium* spp., *F. oxysporum*, *F. solani*, *Pythium* spp., and *P. ultimum* detected in soil samples from sites A (columns without pattern) and B (columns with slashes) in 2018 (soil 2018) and 2022, 5 years after the soil treatments of the present study. The intensity of the DNA hybridisation signals is proportional to the amount of DNA in the soil samples: 0 (not detected) to 6 (strongest). The numbers in the columns indicate the signal intensity for each detected fungus within the respective sample.

#### 4. Discussion

A lack of cultivable land for asparagus highlights the need to overcome the asparagus replant problem. While numerous studies have reported around biotic additives as potential biocontrol agents to improve the quality of soil [2,6,29], the transition from theory to practice is insufficient. The present study presents practical findings from field work, to support asparagus farmers.

Soil analyses from the two tested field sites indicated an abundance of different micro-organisms. By means of a DNA Multiscan assay, we detected *Fusarium* spp., *F. oxysporum*, *F. solani*, *Pythium* spp., and *P. ultimum*, of which some are known soil-borne pathogens, at both sites. Since we did not perform more specialised molecular and pathogenicity tests for single micro-organisms, the presence of *Fusarium* spp. and *F. oxysporum* cannot be determined as plant-pathogenic contamination. Among *Fusarium* spp. and *F. oxysporum*, a variety of strains is ubiquitously present in the soil. Most survive either as saprotrophs or as commensal root endophytes. Aside from not affecting plant fitness, they can even protect from disease caused by vascular pathogens [30,31]. Strains of non-pathogenic *F. oxysporum* have also been shown to increase yield and constrain disease [16,32,33]. Notably, the aforementioned fungi were still detected at the end of the experiment. However, most of them, including plant pathogenic *Fusarium oxysporum*, *F. solani*, *Pythium* spp., and *P. ultimum*, were less abundant, particularly at site B. Here, the soil treatment with clay minerals had

the highest impact on suppressing the fungi. This may be due to the high sand content and/or water shortage in the soil of site B. Surprisingly, no *F. oxysporum* was found at site A before the start of the trial in 2018. This may be due to the five-year break in asparagus cultivation, during which wheat was grown, likely preventing the levels of soil-borne fungi that are pathogenic for asparagus to sustain themselves. Following the treatment of the soil, the signals for pathogenic fungi were weakest in the sections treated with clay minerals and champost, which showed similar levels. After our five-year study, *F. oxysporum* was detected in all treated soils but was present at higher levels in the untreated control soil. We believe that the pathogenic variety *F. oxysporum* f.sp. *asparagi* was present and inhibited the growth of the plants.

The results of the pot experiments confirm the contamination of the two types of soil. The asparagus plants exhibited restricted growth in the soil from site B, which had a break from asparagus cultivation of only 1 year. These findings demonstrate that soils cultivated with asparagus for a longer period have negative effects on plant growth. Steam sterilisation led to improved root development and total dry biomass in the plants even after only 8 weeks of cultivation, demonstrating that harmful micro-organisms were eliminated by the sterilisation [34]. Our results are in agreement with those of [4] and [8], but also with a study on soil disinfection for apple growth, where the DM of the plants grown in soils treated at 50 and 100 °C was higher than that of plants from untreated soils [19]. The application of clay minerals or root-beneficial micro-organisms was not effective in the pot trials, presumably due to the short assessment period. The biofumigation of soil from site B had the most positive effect on asparagus root and shoot growth compared to steam-sterilisation or champost treatment. López-Moreno et al. [26] also reported the positive effect of biofumigation with *Brassica* pellets on the yield of green asparagus in the first year of harvest.

Our pot trials confirm the statement from [2] that most studies have shown short-term effects. The author also addressed the need for long-term effects to be evaluated in labour-intensive field trials. Therefore, we performed field trials to observe the impact of soil supplements on asparagus growth and yield over 5 years, covering 2 years of plant growth and 3 years of harvesting at two sites that had asparagus as preculture. The selected supplements were expected to counteract soil fatigue by shifting the balance of microbes towards non-pathogenic or beneficial micro-organisms, increasing soil fertility and, thus, microbial activity and improving the soil physicochemical properties. We did not focus on single soil-improving effects but observed the overall impact on plant yield and growth over 5 years. The yields from site A were higher than those from site B. Based on the results of the pot trials and DNA Multiscan analysis, we inferred that site B, on which the asparagus cultivation break was only 1 year, was more heavily contaminated with soil-borne pathogens that directly affected asparagus growth. Our findings contradict those of [5], in which the authors concluded that asparagus yield decreases with increasing duration of the asparagus cultivation break. As observed from the DNA Multiscan assay, *F. oxysporum* was already present at high levels in this soil in 2018 and likely had a negative impact on yield regardless of treatment. Another reason for poor yields at site B was the frost damage in 2019 during the early developmental stage of the plants. It is possible that the plants had still not fully recovered from this after 2 years. Furthermore, the extremely sandy composition of this soil leads to rapid drying during periods of low rainfall, which is detrimental to plant growth.

When compared with the untreated control soil, the total marketable yield from site A was higher for the biofumigated and champost-supplemented sections. The yields from the Micosat- and clay mineral-treated soils were not significantly higher than that of the untreated soil. Moreover, in the 2 harvesting years of site B, the yields from the treated soils were numerically higher (except for the 'Ravel' cultivar in the biofumigated, clay mineral-treated, or Micosat-supplemented soils) but not significantly so when compared to that from the untreated control soil.

We deduce that the impact of soil supplements on plant growth is at its peak in the first few years following application and then diminishes. This conclusion stems from the harvest of the trial's final year (2022) at site A, where no significant differences were observed between yields from the treated and untreated soils. However, since experimental error increases with the number of years, it becomes more difficult to detect the differences. At site A, the effect of biofumigation on marketable yield decreased gradually from 24 to 18 to 12% in the years 2020–2022 when compared to the yield from the control section. Similarly, the yields from the Micosat-treated sections declined from 18 to 13 to 9%. This trend was not observed for the champost treatment since the yield was recorded as 14, 11, and 16% more than that of the control soil, respectively. When comparing the total yields from all 3 years combined, biofumigation, champost treatment, and Micosat treatment yielded 16, 14, and 12% more than the control soil, respectively. From each individual year, the marketable yield of asparagus from the biofumigated soil was not significantly different to the yield from the soils supplemented with Micosat or champost. It is evident that the marked positive effect of biofumigation at site A wore off after only 1 year, while it had no significant impact at site B. Biofumigation has a similar effect to steam sterilisation: it eliminates soil micro-organisms for a limited period (both pathogenic and beneficial), including invertebrates that are important for soil structure [35].

Regarding the effect of clay minerals on plant growth and yield, we argue that larger quantities are necessary to sufficiently impact soils with high sand content. Therefore, further studies are needed to evaluate the ranges of amounts of clay minerals in soils to shed more light on the real impact on the asparagus replant problem in sandy fields. More research on longer harvesting periods is also necessary for biofumigation, champost, and supplementation with Micosat. Further practical studies should also investigate the repeated supplementation of soil with, e.g., different types of compost and beneficial micro-organisms. This could be implemented while preparing the asparagus ridge and enhance its impact, as compared to a single application.

Of the tested biotic and mineral soil additives in the present study, Micosat was the one with the lowest associated cost. Biofumigation was the most expensive due to the purchase of *B. juncea* seed meal and the plastic film necessary to cover the soil surface. The high-cost factor for champost was the transportation of large volumes. The cost of application of clay minerals was moderate.

Of the asparagus variants, 'Gijnlim' remains the most commonly used in Germany. The favourable traits of 'Gijnlim' and 'Ravel' are an early harvest, a high number of spears, and a moderate spear diameter. 'Ramires' produces a larger spear diameter and exhibit a slightly later start of harvest. Its marketable yield was significantly lower than that of 'Gijnlim' on both sites for both the first year and the combined total of all harvest years. This is due to the later harvest start of 'Ramires'. A longer harvest period in the following years could compensate for this disadvantage.

## 5. Conclusions

The impact of soil supplements appears to be site-specific, likely dependent on soil properties, pathogen abundance, and environmental conditions. The short-term effects of additives could not be detected in the pot experiments, except for biofumigation. Based on previous studies and the present field experiment, a single overarching solution remains elusive, but a complex combination of measures may sustain the long-term productivity of asparagus.

The biofumigation of the soil in the field study appeared to be the most effective measure initially, increasing the asparagus yield by 16% when compared to the untreated soil. However, this was only true for the site with a lower impact on plant growth due to low pathogen abundance, as suggested by the pot experiments and DNA Multiscan analysis. Moreover, biofumigation is a relatively costly process, and its effect wears off over time. When comparing the impact of the remaining applications, champost may be the most promising amendment for soil in which asparagus is cultivated.

**Supplementary Materials:** The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/agronomy13061464/s1>, Table S1: Marketable yield (dt/ha) and percentage increase (%) in the ‘Gijnlim’, ‘Ravel’ and ‘Ramires’ asparagus variants in field trials across (A) 3 harvest years from site A and (B) 2 harvest years from site B. The soil treatments included champost, clay minerals, mustard meal (biofumigation), and asparagus root treatment with Micosat; Table S2: Marketable yield (dt/ha) of the ‘Gijnlim’, ‘Ravel’, and ‘Ramires’ variants grown in the field trials across all treatments. Different letters indicate significant differences among the cultivars in each year.

**Author Contributions:** Conceptualization, C.F.; C.F., F.H. and R.D.F.-K. performed the experiments and analysed the data; writing—original draft preparation, R.D.F.-K. and C.F.; writing—review and editing, R.D.F.-K., C.F. and F.H. Project administration, C.F. All authors have read and agreed to the published version of the manuscript.

**Funding:** This research was funded by the European Innovation Partnership for Agricultural Productivity and Sustainability (EIP-agri; grant no. FKZ: 204016000008/80168355) and the Ministry of Agriculture, Environment and Climate Protection Brandenburg (MLUK).

**Data Availability Statement:** The data presented in this study are available on request from the corresponding author.

**Acknowledgments:** The authors thank Anja Müller and Christian Jorzig for their valuable work on the pot trials, as well as Simone Starke and the gardeners at the Leibniz Institute of Vegetable and Ornamental Crops (Grossbeeren, Germany) for their help in setting up the experimental area.

**Conflicts of Interest:** The authors declare no conflict of interest.

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