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Biodegradable microplastics impact on soil: how poly-3-hydroxybutyrate alters microbial diversity and nitrogen mineralization processes

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Abstract

Background Poly-3-hydroxybutyrate (P3HB) is a biodegradable plastic that may affect soil quality and plant growth. To explain the observed deterioration of plant growth, this study investigated the effects of P3HB microplastics on the soil microbiome and its activity related to content of nutrients and their transformation processes. A pot experiment was conducted using soil contaminated with five different doses of P3HB, both with and without maize. Soil mineral nitrogen forms, microbial properties as well as plant biomass were determined.

Results P3HB significantly altered soil properties by stimulating microbial respiration, enhancing carbon turnover, and shifting nitrogen forms, notably reducing NO_3^- availability. The fungal community was more sensitive to P3HB compared to the bacterial one. Fungal genera such as *Tetracladium*, *Exophiala*, and *Pseudogymnoascus* were stimulated; others such as *Gibberella* and *Gibellulopsis* declined. In the bacterial community, P3HB promoted the growth of copiotrophic P3HB degraders (e.g., *Actinobacteria*, *Alphaproteobacteria*); increased the abundance of anaerobes (*Clostridia*); decreased nitrifying groups (*Nitrososphaeria*, *Nitrospiria*); and reduced oligotrophic taxa (*Vicinamibacteria*, *Thermoleophilia*). These changes led to altered nutrient cycling, including inhibited nitrification and reduced mineral nitrogen availability, contributing to decreased maize growth.

Conclusions Soil contamination with ≥ 1% P3HB microplastics disrupts microbial structure and nutrient dynamics, with potential negative effects on soil fertility and plant productivity.

Keywords Biodegradable plastics, Bacteria, Fungi, Nitrification, Soil nitrogen

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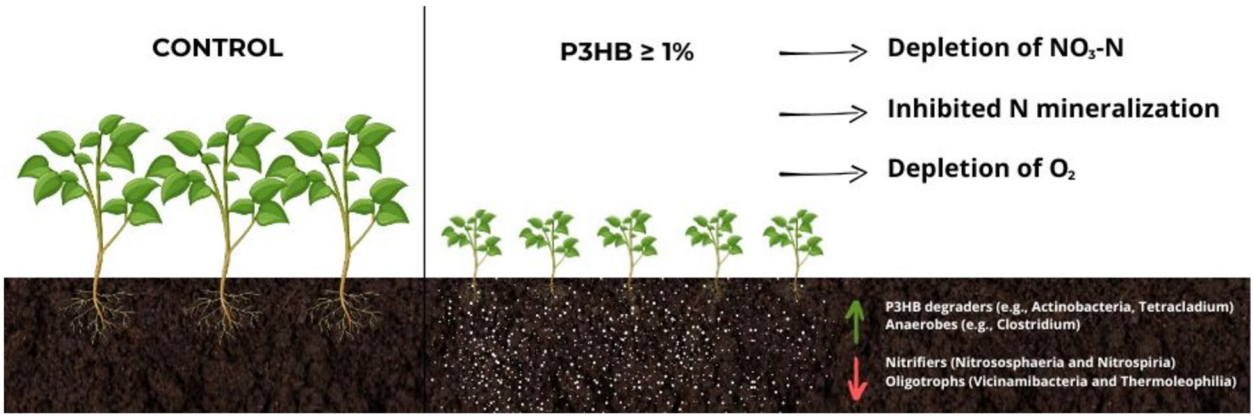
Graphical abstract



Background

Plastic pollution is among the most pressing global environmental threats. One promising solution is the replacement of conventional plastics with biodegradable alternatives. Polyhydroxyalkanoates (PHAs) are a group of natural biodegradable plastics (BPs) valued for their favorable physicochemical and mechanical properties, as well as their ability to undergo degradation under both aerobic and anaerobic conditions [1, 2]. Among them are the most commonly used polyhydroxybutyrates (PHBs), including poly-3-hydroxybutyrate (P3HB) and poly(3-hydroxybutyrate-co-3-hydroxyvalerate) (PHBV). Both polymers are biosynthesized from various carbon (C) substrates by bacteria under nutrient-limited conditions [3]. PHBV can only be biosynthesized by microorganisms in the presence of specific substrates, and it is not commonly found in nature without engineered or optimized conditions. On the contrary, P3HB is the most common and simplest form of PHAs, serving as an intracellular storage compound for C and energy in many types of bacteria [4]. Nevertheless, the increasing application of PHAs in agriculture raises concerns, as residual concentrations in soils may exceed 1.5% [4]. Such a high concentration of BPs could affect agroecosystem functioning.

To date, several adverse effects of PHAs on soil have been reported (Table 1). Since 2021, studies have increasingly documented their presence and progressively revealed potential impacts, particularly on the soil

Table 1 Potentially adverse effects of PHA types in soil

Effect	PHB	P3HB	PHBV	References
Acidification			•	[8]
		•		[21]
Modification of SOM* structure and stability**		•		[22]
Decrease in SOM water binding**		•		[22]
Enhanced SOM degradation coupled with C and nutrient turnover			•	[6, 8]
		•		[4, 23, 24]
Decrease of NO_3–N**	•			[25]
Decrease of available NO_3^- and NH_4^+ in soil solution			•	[6]
Change in the nutrient balance			•	[26]
		•		[21]
Increase in lead (Pb) availability**	•			[25]
Soil microbial community change		•		[23, 24]
			•	[6–8]

*SOM = soil organic matter; ** effect of P3HB microplastics

microbiome. This leads to a debate whether PHAs should be considered as a short-lived (temporarily affecting the environment) contaminant.

Soil microorganisms may be affected by various PHA-induced changes in the soil environment (Table 1). These effects are often accompanied by substantial shifts in both the taxonomic and functional diversity of the soil microbiome [5–8]. The bacterial community tends to be enriched with PHA-degrading taxa such as *Alphaproteobacteria* [9, 10], *Actinobacteria* [4, 11], and presumably also *Gammaproteobacteria* [12] and *Firmicutes* [13] reflecting a broader shift from oligotrophic (slow-growing microbes adapted to low-nutrient environments) [14] to copiotrophic (fast-growing that thrive in nutrient-rich environments) [15] microbial communities. In addition, changes in nitrogen (N) availability affected nitrifying prokaryotes [16, 17]. In addition, several fungal taxa, particularly from the phyla *Ascomycota* [17–19], *Basidiomycota* [18] and *Deuteromycetes* [20] have been identified as PHA degraders and may play a key role in their decomposition.

Although the direct effects of PHAs on soil microorganisms may be considered neutral or even beneficial in some cases, the associated ecological shifts can have negative consequences for the broader soil environment. These include imbalances in nutrient transformation rates and reduced nutrient availability [23], which have been linked to adverse impacts on plant growth [6, 23, 27]. Notably, no phytotoxic effects have been observed in aquatic plants, at least in the case of P3HB [28]. Still, critical questions remain, including the underlying mechanisms driving these changes, type of alterations occurring in the soil microbiome and also PHAs threshold concentrations.

Building on previous works [4, 21–23, 28], this study focused specifically on P3HB and its impact on soil health. A major negative effect of P3HB on plants has been attributed to disruptions in nutrient balance and availability. P3HB serves as an easily utilizable C source, affecting the accessibility and use of native soil organic matter (SOM) by soil microbes [23]. In fact, C represents 56% of P3HB molecule, with no N, thus addition of a small amount of P3HB represents a significant increase of C in a mineral soil. This additional C source alters the soil C:N ratio, pushing it beyond the optimal range (C:N = 7–8.6) for microbial growth [29, 30]. The elevated C:N ratio increases microbial demand for N, leading to N mining from SOM via upregulated N-hydrolase production to restore stoichiometric balance [31]. As a result, rapid microbial utilization of P3HB accelerates metabolic activity and growth and intensifies the nutrient uptake [8, 23]. This action likely depletes N reserves needed for plant development [6, 23, 32]. Although this hypothesis has already been postulated, no previous studies have directly examined N form concentrations in P3HB-contaminated soils.

The effect of polyalkenoates biodegradation of P3HB on plant growth are complex, primarily due to the nutrient competition and microbial community shifts. The microbial proliferation is stimulated by the labile C input, leading to increased microbial respiration and nutrient immobilization. This results in reduced N availability for plants, as microbes outcompete roots for essential nutrients, such as NO_3^- and NH_4^+ [33]. In maize, this effect is manifested as a drastic reduction in above-ground biomass at ≥ 1% P3HB concentration, despite increased microbial biomass C (MBC) and enzyme activity, highlighting a trade-off between microbial stimulation and plant productivity [33]. Similarly, in lettuce, P3HB addition significantly suppressed shoot and root biomass across various soil-to-sand ratios, even when soil N levels were elevated, suggesting that nutrient competition and rhizosphere interactions play a key role in mediating these effects [33].

In some cases, P3HB also altered soil physicochemical properties, including pH decrease, increased C:N ratios, and changes in soil moisture and oxygen availability, which can further limit nutrient uptake by plants [34]. Moreover, the formation of the plastisphere shifted microbial community composition toward taxa specialized in plastic degradation, sometimes at the expense of plant-beneficial microbes [8]. These microbial shifts suppressed nitrification, increased SOM degradation, and triggered additional stoichiometric imbalances in C:N:P dynamics, as demonstrated also in experiments with PHBV [6].

Despite growing interest, few comprehensive studies have explored how P3HB affects the soil microbiome in detail. Current evidence suggests that P3HB microplastics shift soil nutrient dynamics away from C, N, and other nutrient sequestration into SOM, toward their accelerated consumption, especially N [23]. These changes likely coincide with alterations in microbial activity, diversity, and metabolism, ultimately hindering plant nutrition and growth.

However, important knowledge gaps remain. Specifically, little is known about:

I. How P3HB microplastics reshape microbial community composition in relation to microbial activity; and
II. How these changes affect nutrient content and transformation, contributing to reduced plant growth.

This study was designed to address these gaps by testing the effects of different P3HB concentrations on soil. The following hypotheses were proposed:

1. Increasing P3HB concentrations reduce microbial diversity in soil.
2. Input of labile P3HB increases copiotrophic P3HB degraders while reducing nitrifiers and oligotrophs.
3. P3HB degradation enhances microbial N acquisition, leading to soil N depletion and reduced plant-available N.

Materials and methods

Experimental design and sampling

The soils used in the pot experiment were prepared by mixing arable soil, sieved through a 2 mm mesh, with P3HB powder (< 80 µm particle size) in five variants based on the proportion of P3HB: 0, 0.1, 1, 5 and 10% w/w. In agricultural soils with P3HB mulching sheets, P3HB concentrations typically range from approximately 0.5–1.5%, but are expected to increase in the future [4]. Accordingly, the tested P3HB concentrations (c(P3HB)) were categorized as low (0.1%), medium (1%), and high (5 and 10%), with the 0% variant serving as the control.

The arable soil used was classified as a silty clay loam (USDA Textural Triangle) Haplic Luvisol (WRB soil classification) sampled (0–15 cm) near the town Troubsko, Czech Republic (49°10′28″N 16°29′32″E). The chemical properties of the soil were: total C 14.0 $g{\cdot}kg^{-1}$, total N 1.60 $g{\cdot}kg^{-1}$, P 0.10 $g{\cdot}kg^{-1}$, S 0.15 $g{\cdot}kg^{-1}$, Ca 3.26 $g{\cdot}kg^{-1}$, Mg 0.24 $g{\cdot}kg^{-1}$, K 0.23 $g{\cdot}kg^{-1}$; pH ($CaCl_2$) 7.3. P3HB powder (ENMAT Y3000) from TianAn Biologic Materials Co., Ltd. (Ningbo City, China) was used for this experiment. P3HB is slightly hydrophobic polymer having the contact angle between 70° and ≈81°. Further specification of used P3HB can be found in the study of Fojt et al. [22].

Experimental plastic pots (2 L) were filled up with 1.7 kg of the respective soils. Each treatment was prepared in 10 repetitions (pots). Then, five pots of each treatment were sown with maize seeds (*Zea mays* L.) and five were run without plants. In fact, five seeds were sawn and after sprouting in the pot, only two plants were left and then used in the experiment. All experimental pots were placed randomly into growth chamber (CLF Plant Climatics GmbH, Germany), where controlled conditions were maintained: 12 h long photoperiod, light intensity 20 klx, temperature (day/night) 20/12 °C, relative air humidity (day/night) 45/70%, moisture level 65% of water holding capacity. Soil moisture was maintained at this level by watering every other day. After 90 days, tested plants were harvested at ground level, their height was measured and then the plant biomass was dried at 60 °C to the constant weight to determine the yield of above-ground biomass (AGB) in dry weight from each pot. Furthermore, soil samples were taken from all experimental pots.

Soil chemical and biochemical analyses

The soil samples were taken as composited from three probes per each pot, homogenized by sieving through 2 mm mesh sieve and directly used (fresh samples), stored at 4 °C (in refrigerator) or freeze-dried and stored at − 20 °C (in a freezer).

The fresh samples were used for determination of mineral N, NO_3^- (mercurous sulphate electrode type RME 121; Monokrystaly Turnov, Czech Republic) and NH_4^+ (with ammonia gas electrode type 10–23; Monokrystaly Turnov, Czech Republic) [35]. The N (mineral, ammonium, nitrate) content was calculated to the dry soil mass; the dry mass was determined gravimetrically (on laboratory scales) after drying of soil in the laboratory dryer at 105 °C to the constant weight [36].

The fridge-stored samples (at 4 °C) were used for soil respiration analyses. Soil respiration induced by citric acid (Cit-IR), *D*-trehalose (Tre-IR), *N*-acetyl-*β*-*D*-glucosamine (NAG-IR), *L*-arginine (Arg-IR) was measured using MicroResp® device (The James Hutton Institute, Scotland) and spectrophotometric measurement (Tecan Infinite 200 PRO; Tecan Trading AG, Switzerland) of chromogenic indicator (cresol red) for CO_2 emission in the form of agar–agar gel in the 96-well microplate [37]. Freeze-dried samples were used for *N*-acetyl-*β*-*D*-glucosaminidase (NAG) activity assay, based on the spectrophotometric measurement (Tecan Infinite 200 PRO) of the product (4-nitrophenol, PNP) in the reaction with PNP-derivate of the natural enzyme substrate [38].

Soil biological analyses

DNA extraction from freeze-dried samples was done to carry out soil biological analyses. DNA was extracted from 0.5 g of freeze-dried soil samples using the E.Z.N.A.® Soil DNA Kit (Omega Bio-tek, USA). Isolated DNA was quantified using Nanodrop One (Thermo Scientific, USA) and used to determine the soil microbial abundance as well as diversity and composition of soil microbiome.

Abundance of soil microbiome

The SYBR-Green platform was used on a CFX96 Real-Time PCR detection system (Bio-Rad Laboratories, USA). Real-time PCR was performed to quantify genes amoA (coding for ammonium monooxygenase) to determine ammonia-oxidizing bacteria (AOB) and gene phaZ (coding for P3HB depolymerase) to determine generally

all microbial P3HB degraders (phaZ) biomass in soil DNA extracts. The primers used were AMOA1F (5′ GGGGTTTCTACTGGTGGT 3′) and AMOA2R (5′ CCCCTCKGSAAAGCCTTCTTC 3′) for AOB [39]; and PHBf (5′ CGTCTACCGCAACGGCACCAAGG 3′) and PHBr (5′ TGGGCGTAGTTGCTGGCCGT 3′) for phaZ [40].

Diversity and composition of soil microbiome

The structure and composition of the prokaryotic and fungal communities in the soils were analyzed using high-throughput sequencing of fragments the prokaryotic 16S rRNA genes and fungal ITS regions. Specific regions of the rRNA genes of fungi ITS2 (18S) and bacteria V3–V5 (16S) were amplified using primers F357 (5′-CCT ACGGGAGGCAGCAG-3′) and R926 (5′-CCGYCA ATTYMTTTRAGTTT-3′), or ITS3F (5′-GCATCGATG AAGAACGCAGC-3′) and ITS4R (5′-TCCTCCGCT TATTGATATGC-3′), respectively, with barcodes and the universal overhang. Illumina sequencing adaptors were introduced in the second PCR, all in accordance with the general instructions [41]. The products were evaluated by agarose electrophoresis, quantified with a fluorimetric, high sensitivity AccuGreen Quantification Kit (Biotium Inc., USA) and pooled into an amplicon library. Sequencing took place on a MiSeq unit (Illumina, USA) running the reagent kit v2 and paired-end 250 nt reads in an external laboratory (SEQme s.r.o., Czech Republic). After pairing and filtering, a total of 68,025,630 prokaryotic and 2,074,317 fungi reads were obtained and further processed.

Data processing and statistical analyses

The α-diversity of the procaryotic and fungal microbial communities was determined by the Simpson and Shannon diversity indices to reflect changes in the community richness and evenness between experimental groups, and β-diversity was measured by the Sørensen similarity index. While α-diversity is a measure of microbiome diversity applicable to a single variant, β-diversity is a measure of the similarity or dissimilarity of two communities [42].

Data processing and statistical analyses were performed using R, version 4.3.1. [43]. Canonical correspondence analysis (CCA) was carried out to provide advanced analysis of relation between taxonomical β-diversity and key soil nutrition properties and other available parameters. The X axis separated samples according to the added P3HB dose with low c(P3HB) on the left and high on the right. Y axes reflected the presence of maize in the soil with planted variants in the upper and unplanted in the lower part of the scatter plot. The scatter plot was divided into two subplots: one reflected the taxonomic groups (class level) and the other the soil factors (PAST version 4.0) [44].

Principal Coordinate Analysis (PCoA) is multivariate eigen analysis method to explore and to visualize similarities or dissimilarities of data [45]. It converts data on distances between items into map-based visualization of those items, it also allows to identify groups or clusters. PCoA finds the main axes through a matrix and calculates a series of eigenvalues and eigenvectors. Each eigenvalue has an eigenvector, and there are as many eigenvectors and eigenvalues as there are rows in the initial matrix.

For characterization of the relationship among the treatments and selected soil properties, two-way analysis of variance (ANOVA) type I (sequential) sum of squares was used at significance level of 0.05 [46], where the first factor was c(P3HB) and the second factor was presence of maize (unplanted and maize-planted). The purpose of using two-way ANOVA was also testing the interaction between these factors. An effect size was measured using eta-squared (η_p^2) from package lsr [47]. To detect the exact difference among factor level means, it was used Tukey's honestly significant difference (HSD) test from package agricolae [48] and treatment contrast for calculating factor level means with standard error of mean (SEM). These results were graphically represented with bar charts showing statistically significant difference at significance level of 0.05 indicated by different letters. Lowercase letters indicate differences among c(P3HB), uppercase letters between unplanted and maize-planted variants with dependence of P3HB dose.

The data on diversity and composition of soil microbiome were further processed with the DADA2 v1.26.0 R package [49] with help of MetaCentrum high performance computing service (national grid infrastructure of the Czech Republic, which provides researchers, scientists and students with access to powerful computing resources for solving challenging computational tasks) and visualized by the phyloseq v1.42.0 R package [50] and ComplexHeatmap v2.14.0 R package [51]. Package ggplot2 v3.5.1 [52] was used for creating advanced statistical graphs.

Microbiome taxonomy was assigned for the bacteria according to the SILVA 132 SSU NR 99 reference database [53] and the 8.3 release of the UNITE reference database for fungi [54].

Beta diversity among the variants of the experiments (PERMANOVA) was investigated using the vegan R package with Bray–Curtis distance metrics [55]. Random forest statistical method was used to identify potential biomarkers characteristic of the individual variants of the experiment [56]. The results of these analyses are detailed in the Supplementary material.

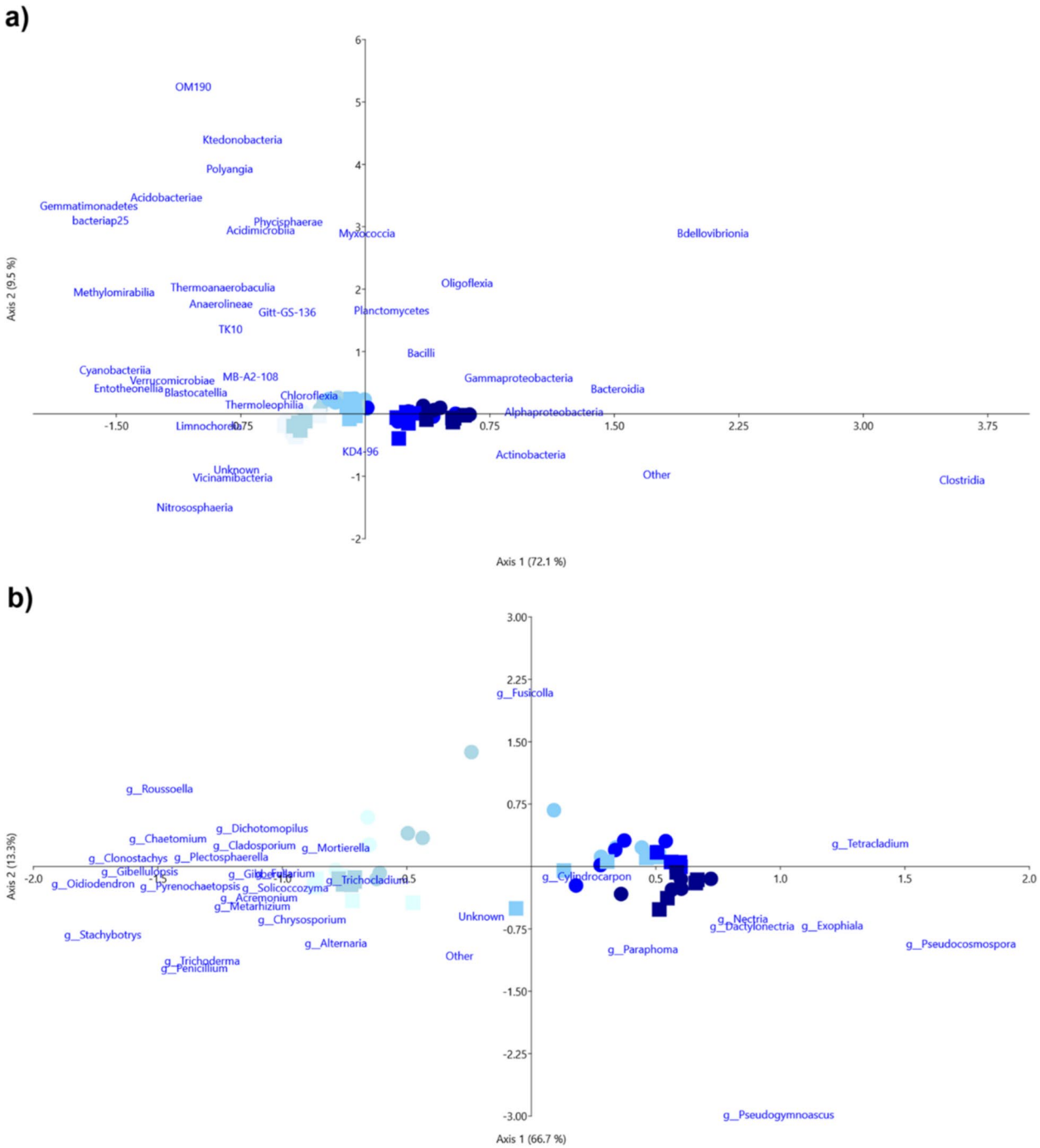


Fig. 1 Canonical correspondence analysis of microbial community composition. Scatter plots showing relations of (**a**) prokaryotic community at the class level and (**b**) fungal community at the genus level in unplanted (square) or planted (circle) soil variants contaminated with variable doses of P3HB (0, 0.1, 1, 5, 10%). Related statistical data are in Tables S1 and S2

Results

Microbial diversity response to P3HB

Despite the differences in relative abundances (RAs) of prokaryotes and fungi (Fig. S1), the Shannon and Simpson indexes showed no significant differences between the prokaryotic communities of the variants (Fig. S2a, b). Similarly, there were no significant differences in fungal communities' diversity in the maize-planted variants (Fig. S2c, d). In the unplanted variants, however, P3HB significantly affected fungal α-diversity: the Shannon index

decreased significantly at 1% c(P3HB) and remained at a similar level for higher c(P3HB) (Fig. S2c). Although the Simpson index showed deviations, no clear trend was observed (Fig. S2d).

Soil taxonomic group's response to P3HB

The changes in soil microbiome composition were comprehensively documented by CCA plots (Fig. 1) and changes in RAs of taxonomical groups, expressed as percentages of all the identified operational taxonomic units found by sequencing.

In the prokaryotic community, increasing c(P3HB) negatively influenced families, such as *Nitrososphaeraceae* and *Vicinamibacteriaceae,* while other families such as *Microbacteriaceae, Caulobacteriaceae, Pseudomonadaceae,* and *Rhizobiaceae* increased in abundance (Fig. S1c). Fungal families *Helotiaceae*, *Herpotrichiellaceae* and *Pseudeurotiaceae* were stimulated by increasing c(P3HB) (Fig. S1d), other fungal families, especially *Nectriaceae* and *Plectosphaerellaceae*, were rather suppressed.

Prokaryotic microbiome analysis showed that *Bacteria* generally dominated over *Archaea*, which were mainly represented by microorganisms involved in nitrification (Fig. S4a). This natural domination was graded in favor of *Bacteria* with increasing c(P3HB) as RAs of *Archaea* decreased (Fig. S4b). Enrichment of the prokaryotic community with *Actinobacteria*, *Alphaproteobacteria*, *Gammaproteobacteria* and anaerobes *Clostridia* was found for high c(P3HB) variants (Fig. 1a). The same figure also shows that control and low c(P3HB) variants were characterized by higher abundance of *Nitrososphaeria*, *Nitrospiria*, *Thermoleophilia* and *Vicinamibacteria*.

CCA plots of both bacterial and fungal taxa (Fig. 1a, b, respectively) showed marked dispersion of variants along X axis, indicating a strong influence of P3HB and clear separation between 0% + 0.1% and 1–10% variants. *Tetracladium*, *Exophiala, Pseudogymnoascus, Pseudocosmospora* and *Nectria* were stimulated by high c(P3HB); these taxa overlaid all other fungi taxa (Fig. 1b).

Actinobacteria, Alphaproteobacteria and *Gammaproteobacteria* represent mostly copiotrophic taxa. As it can be identified in Fig. S6a and Fig. 2, RAs of these prokaryotic groups showed growth following P3HB contamination. *Actinobacteria* was the most abundant prokaryotic group, stimulated by P3HB in both planted and mainly unplanted soil with the highest RA reaching 30 in 10% P3HB maize-planted variant (Fig. 2a). RA of *Alphaproteobacteria* was even more directly dependent

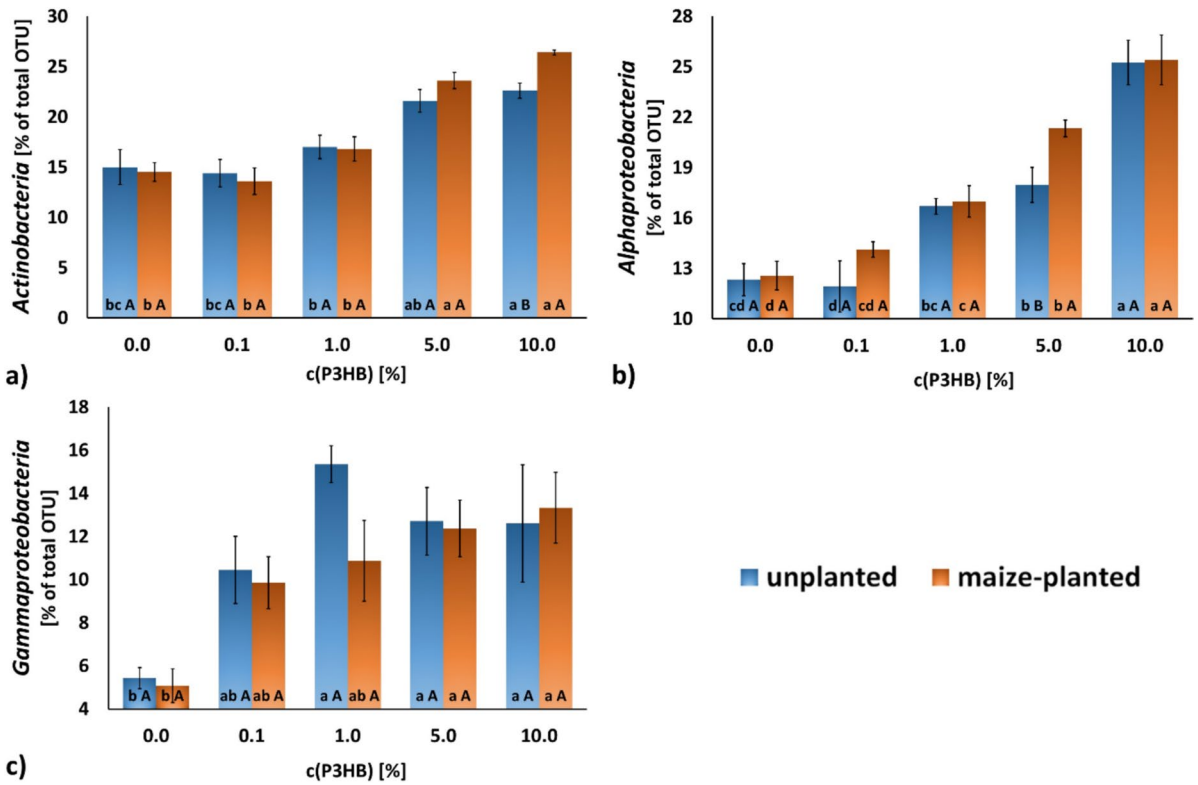


Fig. 2 Relative abundances of the most dominant prokaryotic (bacterial) taxonomic classes in the variants. Average values ($n = 5$) with standard errors of mean (error bars); lowercase letters indicate differences between all the variants (evaluated separately for unplanted and planted variants), uppercase letters between unplanted and planted variant of respective P3HB dose

on c(P3HB) in the soil (Fig. 2b). RA of *Gammaproteobacteria* mostly significantly increased at 1–10% c(P3HB) without another trend (Fig. 2c).

On the contrary, RA of oligotrophic *Vicinamibacteria*, the fourth most abundant prokaryotic group (Fig. S6a), decreased with increasing c(P3HB) (Fig. S5a). Similar trend showed oligotrophic bacteria class *Thermoleophilia* (Fig. S5b).

The RA of groups strongly involved in N cycling, i.e., nitrifying prokaryotic classes *Nitrososphaeria* belonging to *Archaea* and *Nitrospiria*, which oxidize nitrites to nitrates, also decreased as c(P3HB) rose (Fig. 3), mirroring the overall *Archaea* trend (Fig. S4b).

Results in Figs. 1a and S6a indicate that another significantly P3HB-affected class was anaerobic *Clostridia*. Its RA increased significantly at the high c(P3HB) (Fig. S5c), suggesting the development of more anaerobic soil conditions.

Similarly, Figs. 1b, 4a–c, and S7 show that P3HB-stimulated fungal genera *Tetracladium*, *Exophiala, Pseudogymnoascus, Pseudocosmospora* and *Nectria* began to form a dominant fungal group at the high c(P3HB) (S6b). *Tetracladium* became dominating taxa starting at 1% c(P3HB) in both unplanted and planted soil. *Exophiala* had a similar progressive but more gradual development (Fig. 4b). Conversely, *Pseudogymnoascus* responded only at 10% c(P3HB) (Fig. 4c). On the contrary, abundance of *Gibberella* and *Gibellulopsis* as well as other taxa were affected negatively by c(P3HB) (Figs. 4d, e and S6b).

Microbial activity and biomass response to P3HB

All analyzed substrate-induced respirations (IRs) and NAG showed stimulation by increasing c(P3HB), with generally stronger responses at high c(P3HB) (Fig. 5).

Expression of the phaZ gene, encoding PHA depolymerase, was also clearly stimulated at high P3HB doses (Fig. 6a), suggesting active microbial degradation processes. In contrast, the biomass of AOB tended to decrease with increasing c(P3HB) in unplanted soil (Fig. 6b), indicating a possible suppression of nitrifiers under these conditions.

Soil N response to P3HB

In the unplanted soil, the NH_4–N content showed a Gaussian function-shaped trend with maximum value observed at 1% P3HB (Fig. 7b). Conversely, NO_3–N (Fig. 7a) and mineral nitrogen (N_{min}) content in unplanted variants (Fig. 7c) decreased progressively with increasing c(P3HB), indicating potential suppression of nitrification or enhanced immobilization.

Effect of P3HB on maize plant growth

AGB of maize significantly decreased reaching moderate c(P3HB) in soil (Fig. S8). This content was decisive for suppressing the growth of maize; a further increase in c(P3HB) did not have additional negative effect on biomass production. The decisive effect of P3HB and related microbial activity in the soil suppressing the growth of maize was also highlighted by the CCA plots (Fig. 8) with clear grouping of (I) separated AGB, (II) NO_3–N and (III) mostly clustered remaining characteristics.

Effect of maize on the soil system

The microbial diversity response to P3HB revealed that in the maize-planted variants, no significant differences were observed neither in fungal nor prokaryotic community diversity based on the Shannon and Simpson indexes (Fig. S2), suggesting a stabilizing effect of the plant.

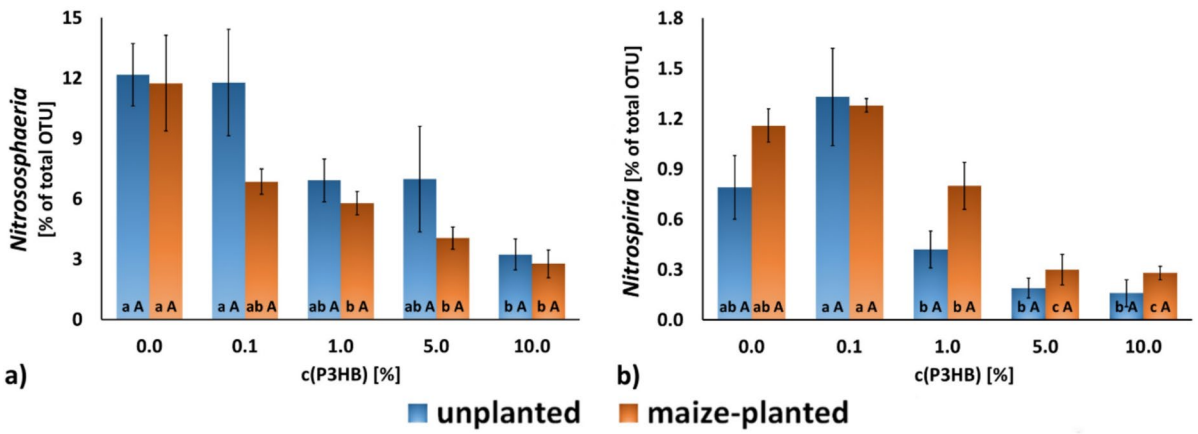


Fig. 3 Relative abundances of nitrifying prokaryotic taxonomic groups in the variants. Average values ($n = 5$) with standard errors of mean (error bars); lowercase letters indicate differences between all the variants (evaluated separately for unplanted and planted variants), uppercase letters between unplanted and planted variant of respective P3HB dose

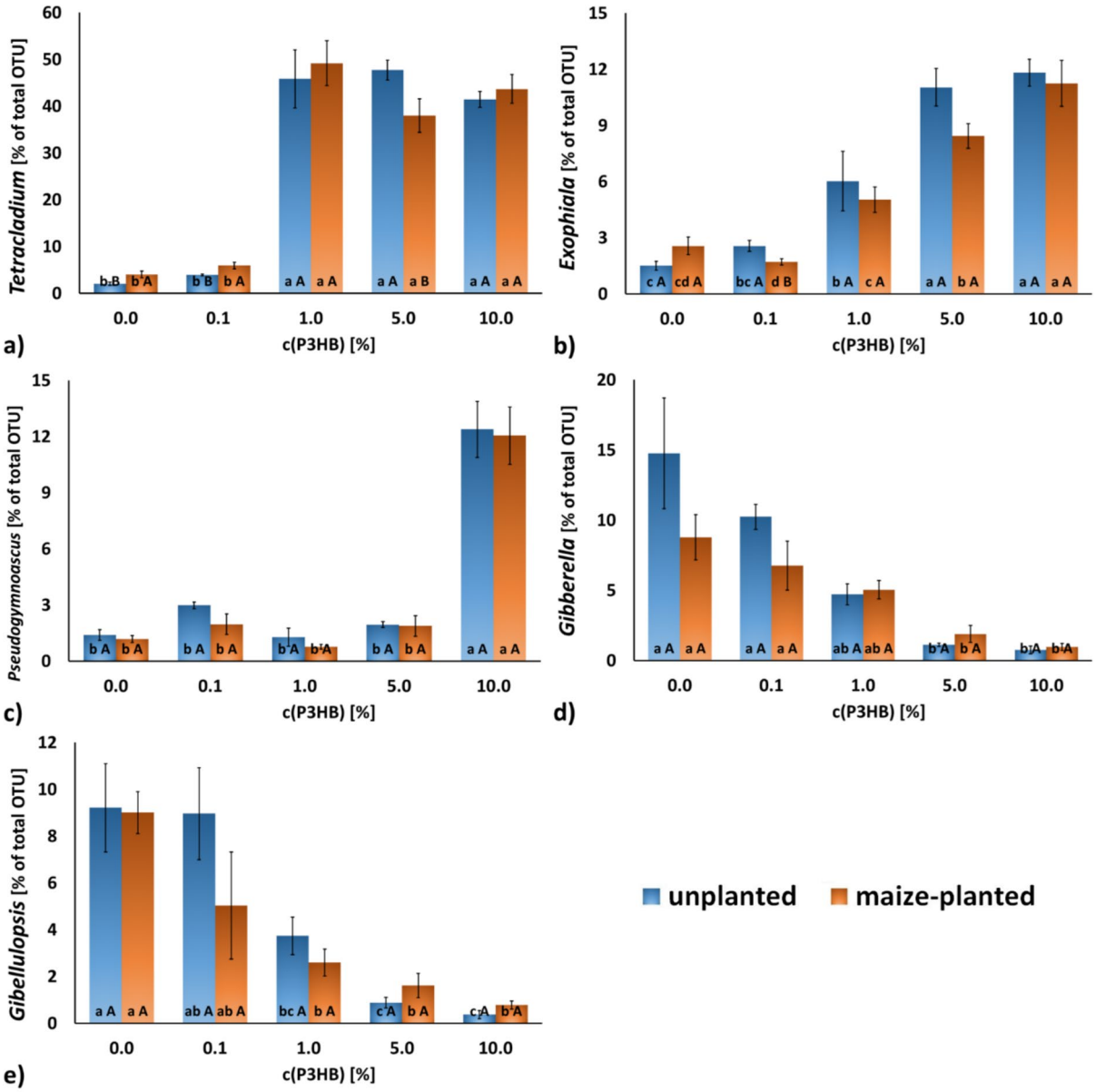


Fig. 4 Relative abundances of fungal taxonomic groups in the variants. Average values ($n = 5$) with standard errors of mean (error bars); lowercase letters indicate differences between all the variants (evaluated separately for unplanted and planted variants), uppercase letters between unplanted and planted variant of respective P3HB dose

PCoA of the prokaryotic community indicated no effect of maize on β-diversity, however, a visible effect of P3HB starting at around 5% c(P3HB) (Fig. S3a). However, PCoA of fungal β-diversity revealed a noticeable influence of plant presence, with clear separation between the control and low c(P3HB) treatments versus the moderate to high ones (Fig. S3b), indicating a combined effect of P3HB and maize on community structure.

Furthermore, the presence of maize had mostly insignificant effects on microbial community structure compared to P3HB (Figs. S1a, b and). Trends observed in the maize-planted variants generally mirrored those in unplanted soil. For example, the dominance of *Bacteria* over *Archaea* increased with c(P3HB), and RA of *Archaea* decreased similarly in both variants (Fig. S4b). The enrichment of *Actinobacteria*, *Alphaproteobacteria*, and *Gammaproteobacteria*, as well as the decline of *Vicinamibacteria* and T*hermoleophilia*, followed the same pattern. Notably, *Actinobacteria* reached the highest RA (30%) in the 10% P3HB planted variant (Fig. 2a).

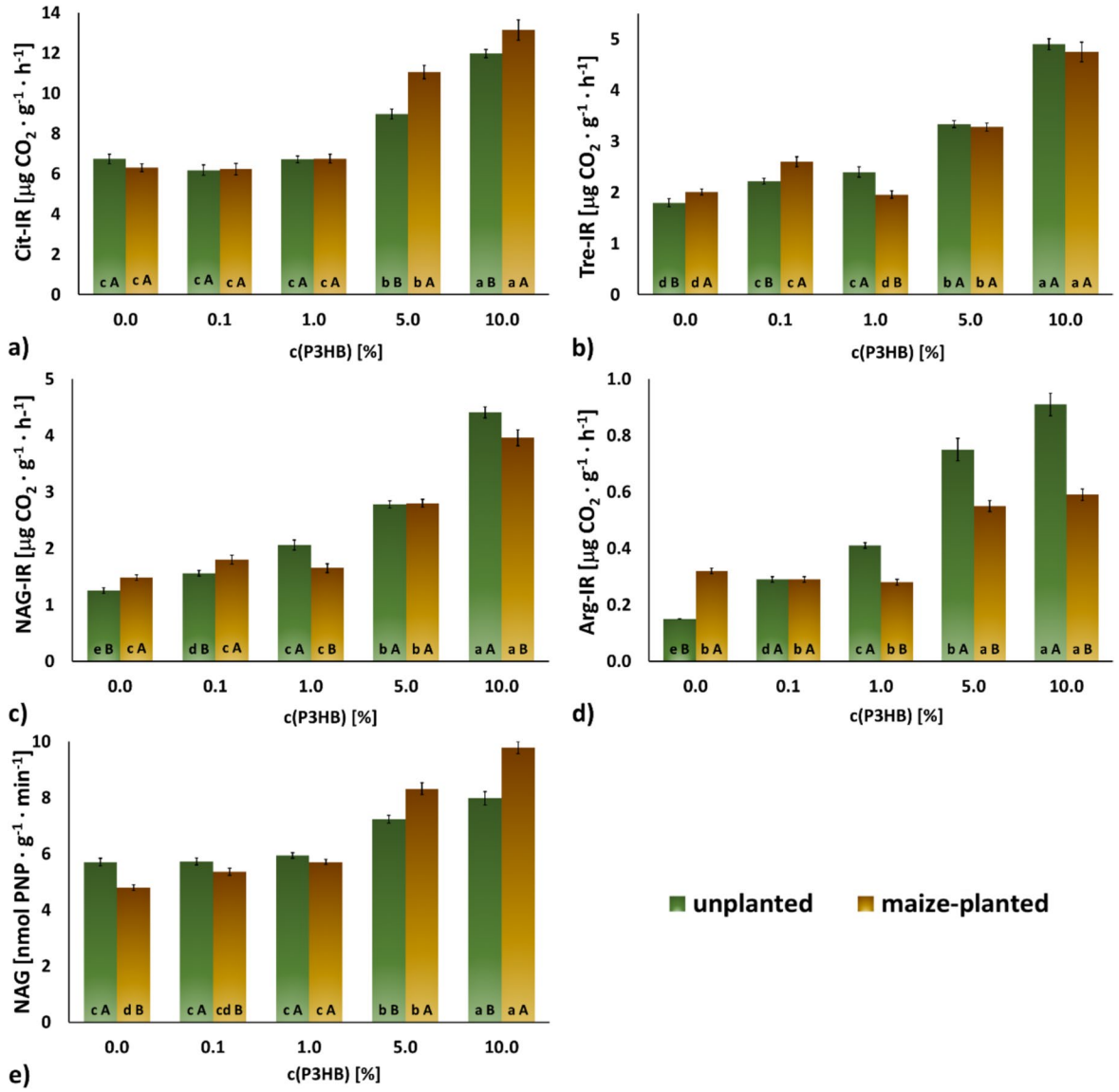


Fig. 5 Respiration induced by substrates—(**a**) citric acid (Cit-IR), (**b**) *D*-trehalose (Tre-IR), (**c**) *N*-acetyl-*β*-*D*-glucosamine (NAG-IR), (**d**) *L*-arginine (Arg-IR)—and (**e**) *N*-acetyl-*β*-*D*-glucosaminidase (NAG) enzyme activity in the variants. Average values ($n = 5$) with standard errors of mean (error bars); lowercase letters indicate differences between all the variants (evaluated separately for unplanted and planted variants), uppercase letters between unplanted and planted variant of respective P3HB dose

Fungal genera such as *Tetracladium* and *Exophiala* also showed comparable responses in planted soil, becoming dominant at similar thresholds of c(P3HB) (Fig. 4a, b). However, for some taxa, such as *Nitrososphaeria* and *Nitrospiria*, the RA decline was slightly steeper in the planted soil (Fig. 3a, b), suggesting that plant presence may modestly amplify the suppressive effects of P3HB on certain nitrifiers. Overall, while maize presence did not significantly reshape the microbial response to P3HB, it may have contributed minor modulations in sensitivity for specific groups.

The analyses of microbial activity and biomass response reported in Fig. 5a–e revealed that in maize-planted soil, the overall trends in substrate-induced respirations and NAG stimulation by P3HB remained similar to those observed in unplanted variants, with increased microbial activity at higher polymer concentrations. However, the presence of maize had a significant effect across all measured characteristics, though without a clear, consistent trend. At lower P3HB levels (0–0.1%), maize appeared to enhance microbial activity, but this effect diminished as c(P3HB) increased. For phaZ (Fig. 6a), a strong

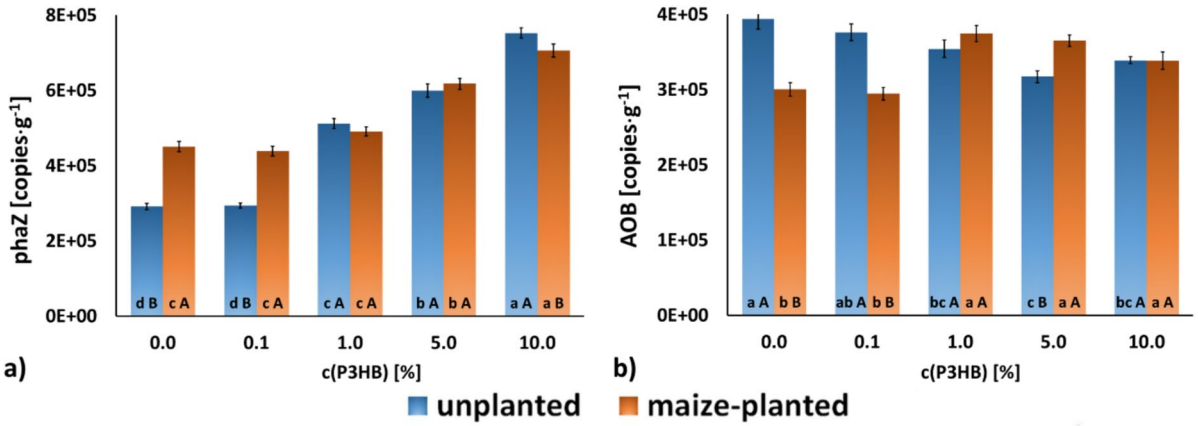


Fig. 6 Microbial abundance of (**a**) P3HB-degrading microbes (phaZ) and (**b**) ammonia-oxidizing bacteria (AOB) in the variants. Average values ($n = 5$) with standard errors of mean (error bars); lowercase letters indicate differences between all the variants (evaluated separately for unplanted and planted variants), uppercase letters between unplanted and planted variant of respective P3HB dose

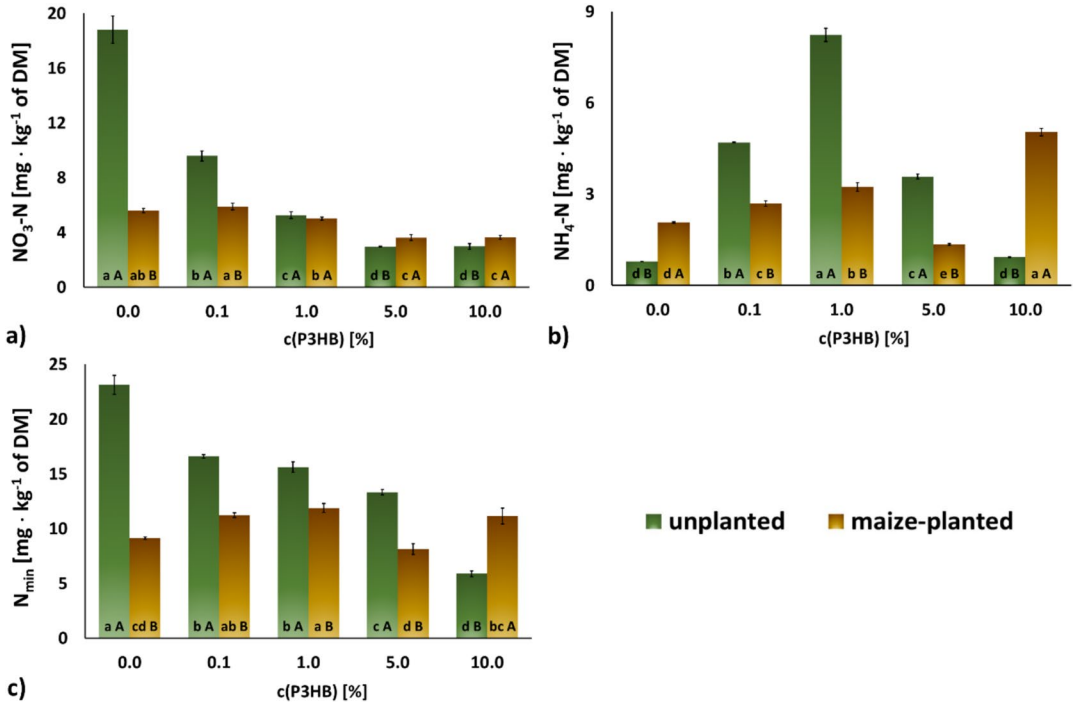


Fig. 7 Content of inorganic N forms in soil. Soil content (in dry matter = DM) of (**a**) nitrate–nitrogen (NO_3–N), (**b**) ammonium–nitrogen (NH_4–N) and (**c**) mineral nitrogen (N_{min}) in the variants. Average values ($n = 5$) with standard errors of mean (error bars); lowercase letters indicate differences between all the variants (evaluated separately for unplanted and planted variants), uppercase letters between unplanted and planted variant of respective P3HB dose

stimulation at high P3HB was observed in both planted and unplanted soils, while the initial positive influence of maize disappeared at higher concentrations.

Interestingly, for AOB biomass, the presence of maize became more relevant: whereas unplanted soil showed a decreasing trend, the planted soil responded positively to P3HB concentrations ≥ 1%, leading to a slight stimulation of AOB biomass (Fig. 6b). Nonetheless, the overall differences remained minor, suggesting that maize modulates but does not override the primary P3HB-driven microbial responses.

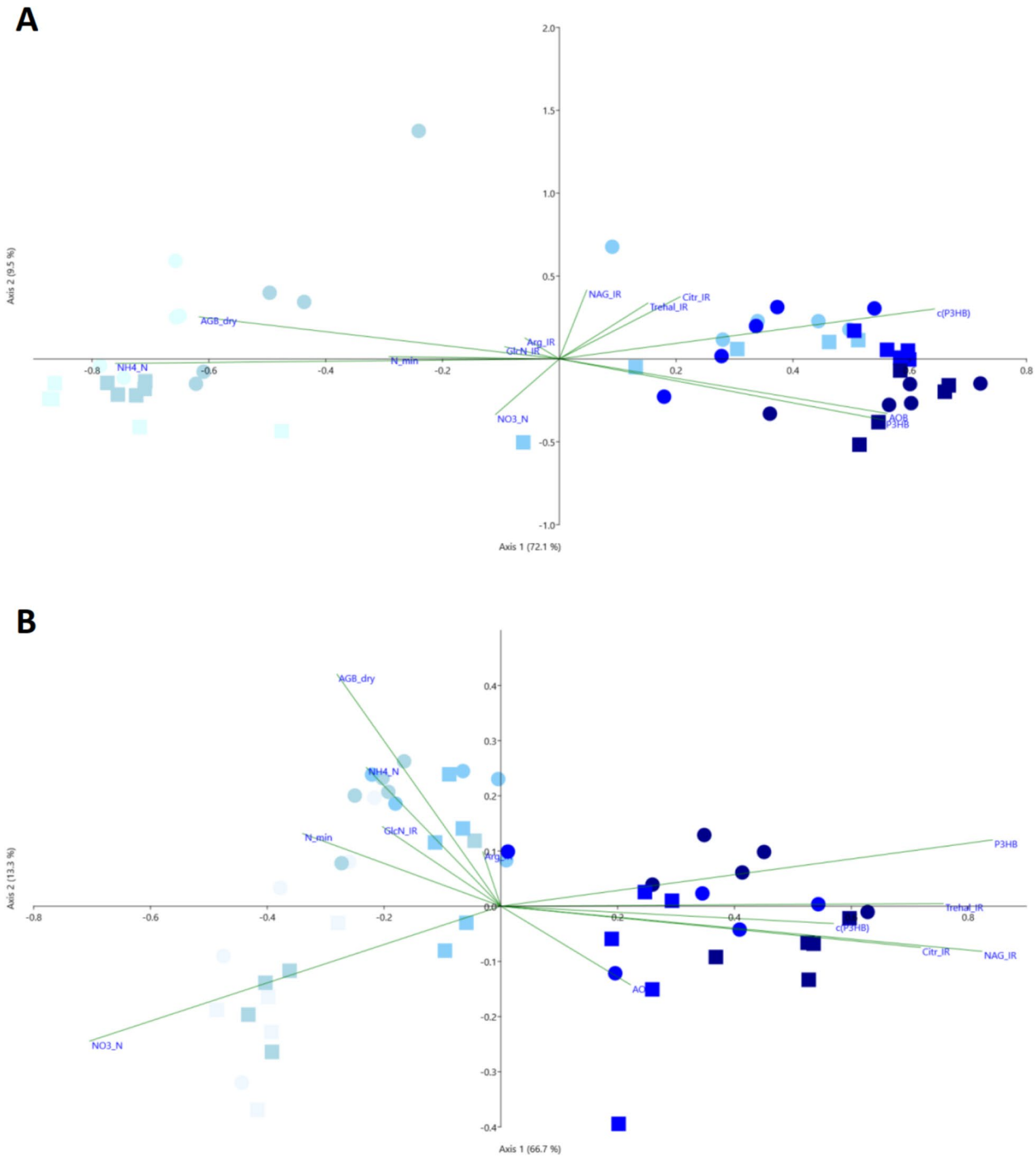


Fig. 8 Canonical correspondence analysis (CCA) of the soil and plant properties. Scatter plots depicting mutual relations of soil chemical (including concentrations of P3HB), microbial and plant (maize AGB dry) properties, determined in unplanted (square) or planted (circle) variants contaminated with variable doses of P3HB (0, 0.1, 1, 5, 10%). (**a**) Bacterial community at the genus level (Axis 1 explains 72.1% of variability; Axis 2, 9.5%) and (**b**) fungal community at the genus level (Axis 1, 66.7%; Axis 2, 13.3%)

The response of NH_4–N to P3HB in the presence of maize showed a positive linear relationship with increasing c(P3HB), except at 5% P3HB, where a deviation from the trend occurred (Fig. 7b). The presence of maize was a statistically significant factor across all treatments. For NO_3–N and N_{min} (Fig. 7a, c), the decreasing trend

observed in unplanted soil was less pronounced. The dependence on c(P3HB) was weaker and mostly indirect, indicating that maize may buffer or modulate the effects of P3HB on N dynamics.

Discussion

Changes in soil microbial community diversity

The unchanged α-diversity of *Prokaryota* (Fig. S2a, b) does not align with previous findings. In soils contaminated with BPs, some authors [8, 57] observed an increase, while other observed a decrease [58]. This contrasts with β-diversity, which showed no effect and an increase at higher P3HB dose (Fig. S3a).

Fungal community diversity was affected by P3HB, as its Shannon index α-diversity decreased at 1% c(P3HB) and higher in unplanted soil (Fig. S2c). In addition, β-diversity was strongly influenced by P3HB (Fig. S3b). In the presence of maize, the Shannon index α-diversity followed the same trend, though the differences were not statistically significant. The stronger fungal response (Fig. 1), evident from the suppression of certain fungal genera compared to prokaryotes, was also observed in the RA of taxonomic groups (Fig. S6b). While most fungal genera were suppressed at moderate c(P3HB) levels, the critical effect on *Prokaryota* occurred starting at 5% c(P3HB). These results indicate a higher sensitivity of fungi to P3HB addition and confirm first hypotheses that increasing c(P3HB) in soil modifies microbial fungal community toward lower diversity.

Moreover, we cannot rule out the hypothesis that the observed decline in fungal α-diversity reflects a loss of generalist species and a shift toward a limited number of P3HB-degrading specialists (e.g., *Tetracladium*, *Exophiala*), which gain a competitive advantage under elevated P3HB concentrations. In this context, the input of P3HB acts as a disturbance, leading to a simplification of the fungal community and promoting specialist taxa equipped with enzymatic capabilities to degrade polyester-type compounds. This supports the ecological theory that disturbances tend to favor specialists adapted to narrow resource niches over generalists [59, 60].

Changes in soil microbial community composition

Both prokaryotic and fungal community compositions were affected by the presence of P3HB in soil (Figs. 1, 2, 3, 4; S4, S5 and S6). The dominance of bacterial RAs compared to *Archaea* increased with higher c(P3HB) (Fig. S4), as the RA of *Archaea* decreased (Fig. 2a). Despite their ability to synthesize and metabolize PHAs [61], soil *Archaea* are more involved in nitrification [62], a process heavily altered by P3HB contamination.

Beta diversity analysis (PERMANOVA) supported these findings (Text S3), showing that experimental variants explained 18.5% of the variance in bacterial communities (Table S5) and 68.1% in fungal communities (Table S7), with P3HB concentration emerging as the main driver of community structure. Pairwise comparisons revealed that microbial composition significantly differed between variants with distinct P3HB concentrations, while the influence of maize alone was minimal or limited (Table S6 and S8). These results were corroborated by Random Forest analysis (Text S4), which identified multiple microbial biomarkers associated with specific P3HB treatments (Fig. S10). No such biomarkers were detected when grouping by maize presence, further confirming the dominant effect of P3HB over plant presence on microbial composition. Some of the previously highlighted abundant taxa, such as *Tetracladium* and *Actinobacteria*, were identified as group-specific indicators, alongside other low-abundance taxa less visible in RA-based summaries (Text S4 and Fig. S10).

Highly P3HB-stimulated copiotrophic classes *Actinobacteria, Alphaproteobacteria* and *Gammaproteobacteria* (Figs. 2 and S6a) likely comprised the majority of prokaryotic P3HB degraders (phaZ gene) in this experiment (Figs. 1a and 6a). The ability of *Actinobacteria* to degrade P3HB is well-documented [63] and is often coupled with their potential to synthesize and accumulate P3HB [64], with genera such as *Streptomyces*, *Rhodococcus*, and *Microbacterium* reported as active degraders. Within *Alphaproteobacteria,* genera such as *Cupriavidus* and *Beijerinckia* are known to produce and degrade PHAs [65, 66]. *Gammaproteobacteria,* including *Pseudomonas*, *Comamonas*, and *Aeromonas,* also harbor PHA depolymerases and were likely involved in P3HB degradation under enriched conditions.

Increasing c(P3HB) led to the formation of a dominant fungal group consisting of the genera *Tetracladium*, *Exophiala* and *Pseudogymnoascus*, with weak enrichment in other known and unassigned fungi (Fig. S6b). This finding confirms the existence of an abundant P3HB-degrading fungal population in the soil. The increased aerobic mineralization of C sources (Fig. 5b, d), representing products of fungal growth, metabolism, and biomass turnover, indicated a strong fungal involvement in P3HB degradation. The booming *Tetracladium* from *Ascomycota* group might be the most actively involved.

Tetracladium species, such as *Tetracladium marchalianum,* can degrade carboxymethylcellulose, xylan and polygalacturonic acid [67]. *Tetracladium* genomes were found to contain various esterases, lipases and pectate lyases, which are important enzymes for the degradation of some polyesters, e.g., poly(butylene succinate-co-adipate) (PBSA) [68]. The authors of the aforementioned study reported that fungal communities on PBSA and in the surrounding soil were strongly dominated by

Tetracladium spp. (RA 42–45%), and the PBSA micro-BPs were substantially mineralized to CO_2. Our results (RA up to 50%; Figs. 4a and S6b) are consistent with these findings.

Toxic and extreme-tolerant class *Exophiala,* belonging to the black yeast *Ascomycota* group, also thrived at 1–10% c(P3HB) (Figs. 4b and S6b). Black yeast species, such as *Exophiala oligosperma* R1, *E. xenobiotica* or *E. jeanselmei*, have been shown to degrade various C substrates, e.g., aromatic hydrocarbons or low-molecular-weight urethane compounds [69–72]. Their abilities predispose them to be efficient in BP degradation and support our findings.

The psychrotolerant *Ascomycota Pseudogymnoascus* responded positively to the highest P3HB contamination (Figs. 4c and S6b). *Pseudogymnoascus* species are known to be cellulolytic and function as saprotrophs [73]; they are either psychrophilic or psychrotolerant and abundant in soils with high fertility [74] indicated here by high c(P3HB) and originally fertile arable soil. The involvement of their representatives in BPs degradation was found recently [75].

The negative effects of P3HB on the soil microbiome and the decrease in RAs of groups such as *Thermoleophilia, Vicinamibacteria, Nitrospiria, Nitrososphaeria*, *Gibberella* and *Gibellulopsis* (Figs. 3, 4d, e; S5 and S6) may be associated with related changes in elemental cycles and metabolic conditions for these microorganisms. Thus, the second hypothesis was confirmed.

Changes in soil microbial behavior and processes related to C

The CCA results (Figs. 1 and 8) provided further evidence of changes in the composition and behavior of the microbial community and the association of P3HB degraders with the mentioned stimulated groups of bacteria and fungi. These changes were accompanied by a shift in the substrate consumption preferences from intrinsic soil organic C to P3HB [21, 23], as confirmed by the increase in both the phaZ gene (Fig. 6a) and microbial activity properties (Fig. 5). This shift probably initiated further changes in soil microbial activity, metabolic processes and soil environment.

The results are of particular interest as they do not align with the general understanding of the adaptation of fungi and bacteria to shifts in SOM (i.e., substrate) quality. From a metabolic pathway perspective, bacteria often possess more simplified, direct, and diverse metabolic pathways compared to fungi, enabling faster adaptation to various conditions and stresses. Bacteria produce a wide range of enzymes that allow them to quickly metabolize various substrates quickly, whereas fungi typically have more complex metabolic pathways and rely on extracellular enzymes to break down complex organic molecules. Bacteria reproduce rapidly and can quickly colonize and adapt to new environments, although their growth is often limited by the availability of nutrients. On the contrary, fungal growth and adaptation to stressors are generally slower.

Changes in soil microbial behavior and processes related to oxygen (O)

All the above-discussed leading stimulated taxa represent aerobic microorganisms, while the *Clostridia* class also includes anaerobic species. *Clostridia* proliferation was highly stimulated at high c(P3HB) levels (Fig. S4c); this may indicate a decrease or depletion of O_2 in the soil following the strongly enhanced P3HB mineralization-based respiration (Fig. 5). This factor could have contributed to the deterioration of environmental conditions for other soil organisms. Furthermore, a decrease in O_2 in the vicinity of plant roots could exacerbate the adverse effect of P3HB on plant roots [76]. To support or reject this new hypothesis, a pilot respiration/biodegradation experiment was conducted (Text S1).

The experiment revealed a sharp decrease in O_2 in initial stage of biodegradation (Text S2, Fig. S9). After a few weeks, the O_2 level increased again stabilized, although it was still slightly lower compared to the control. That implies that O_2 concentration fluctuates during the biodegradation process and its consumption for biodegradation is higher in the initial stage of the process. As a result, we conclude that for plant growth in the presence of biodegrading P3HB, the O_2 shortage may be problematic mainly in the early stage of P3HB introduction, where is the rate of biodegradation highest (Text S2). If the initial phase of plant growth occurs simultaneously as in this and other experiments [21, 23], this deficiency can be critical. This aligns to results of Brown et al. [6], who have analyzed stress markers in maize grown in the soil amended with PHBV microplastics. Even at low PHBV concentrations, the authors found significantly higher content of lactic acid (a product of anaerobic respiration), which authors attributed to a response of plant to PHBV-induced hypoxia in the soil.

From the environmental perspective, under strictly anaerobic conditions (e.g., in paddy soils), the enhanced proliferation of anaerobic microorganisms is problematic due to possible proliferation of methanogenic microorganisms and the subsequent production of methane [77], which is a common product of ultimate biodegradation under anaerobic conditions. Under common conditions, ideal for P3HB biodegradation, the level of O_2 may vary, but the conditions may not be supportive for methanogenesis. Nevertheless, as shown by Lussich et al. [78] or discussed by Schlüter et al. [79], due to local O_2 shortage

in otherwise well-aerated soils, microbial activity in hotspots with easily degradable organic compounds and available nitrate (e.g., arable soils) may facilitate denitrification leading to N_2O emissions [80]. Hence, the additional research is needed to shed light on this environmental aspect of P3HB and other BPs biodegradation.

Changes in soil N-mineralization

N-mineralization is a process by which microorganisms in the soil decompose organic N compounds into inorganic forms, primarily ammonium (NH_4^+). This process occurs as part of the decomposition of organic matter and added substrates. It involves several consecutive steps, such as decomposition (microbial breakdown of organic matter), ammonification (conversion of the organic N into ammonium) and, in well-aerated soil, an optional step of nitrification (conversion of ammonium into nitrate NO_3-by nitrifying bacteria) [81]. In line with above discussion, we speculate that the P3HB-induced decrease of AOB in the unplanted soil (Fig. 6b) may be related to the decrease in the RA of *Archaea* (Fig. S4). AOB and ammonia-oxidizing *Archaea* occupy the same ecologic niche, differentiated only by the availability of ammonium in the environment [61, 82]. Ammonia-oxidizing *Archaea,* including *Nitrososphaeria*, are abundant in warm and humid soils, along with AOB (including *Nitrospiria*). Both groups play a significant role in soil nitrification. *Nitrososphaeria* represents a dominant group of ammonia-oxidizing *Archaea* within *Nitrososphaerota* in arable soils [58], while *Nitrospiria* is the key bacterial group for soil nitrification [83]. Therefore, the decreasing RAs of both classes with increasing c(P3HB) (Fig. 3) confirmed changes in N cycling in the soil, which were directly reflected by altered contents of N forms (Fig. 7).

The decrease in the RA of *Archaea* with increasing c(P3HB) can be explained by the addition of a substrate that does not contain N or NH_2 groups. In other words, there is no N-mineralization, and nitrifying organisms are not needed as ammonia or ammonium ions are not released. Instead, other types of organisms proliferate and exploit these new conditions. A predominant decrease of NO_3–N, N_{min} (Fig. 7a, c) and AOB (Fig. 6b) with increasing c(P3HB) in the unplanted soil supported the assumption of a strong negative effect of soil P3HB contamination on nitrification. This effect was modified by the presence of maize, mostly due to competition between the plant and microbes for N acquisition. N uptake by maize decreased NO_3–N and N_{min} in the control (Fig. 7a, c); however, the difference gradually decreased and even reversed with increasing c(P3HB), which may be related to the severely limited plant growth starting at 1% c(P3HB) (Fig. S8).

The preferential boosted utilization of high c(P3HB) as the primary C source in soil, accompanied by increased utilization of N compounds from SOM (Figs. 5d and 6a), enhanced nutrient uptake and catabolism. This likely resulted in a partially anoxic environment (chapter 4.4) with a more severe impact in planted soil due to the parallel root respiration exhausting soil O_2. Partial anaerobiosis could lead to the subsequently inhibited nitrification (Fig. 3) and explain NO_3–N depletion (Fig. 7a), as nitrate could be used as an alternative electron acceptor for P3HB degradation [8, 84]. The same mechanism can explain higher NH_4–N content in the moderate unplanted P3HB variant and rather increasing NH_4–N content in the planted soil (Fig. 7b), because nitrification is prevented by O_2 limitation. As a result, 1% c(P3HB) in the soils (used in this work or similar in terms of SOM content) appears to be a critical breakpoint above which crucial changes occur.

To sum it up, the observed changes in N cycling can be mechanistically explained by the interaction between P3HB degradation and microbial activity in soil. As a labile, N-free polyester, P3HB provides an abundant and readily degradable C source, which strongly stimulates the growth and metabolism of specialized microbial degraders, particularly copiotrophic bacteria. This sudden influx of C disrupts the natural C:N balance in the soil, shifting microbial communities toward increased N demand to support biomass synthesis.

However, since P3HB contains no N or amino groups, its degradation does not contribute to ammonium release through mineralization. In response, microorganisms begin to mine N from SOM, producing extracellular enzymes such as proteases and ureases to access organically bound N. This microbial N-mining process increases the demand for mineral N (particularly ammonium), but due to the lack of N input from P3HB itself, this process soon becomes limited. As a result, the supply of ammonium available for nitrification declines, leading to the observed suppression of AOB and *Archaea*, including key taxa, such as *Nitrososphaeria* and *Nitrospiria*. These groups occupy the same ecological niche but differ in their adaptation to ammonium availability; both play essential roles in nitrification, and their decline reflects a broader disruption in the N cycle. This is supported by the progressive decrease in nitrate and total mineral N (N_{min}) content with increasing P3HB concentrations, particularly in unplanted soils.

Moreover, the rapid microbial metabolism of P3HB likely increases oxygen consumption in the soil environment. In planted treatments, root respiration further exacerbates oxygen depletion, creating micro-anaerobic conditions. These conditions are unfavorable for aerobic nitrifiers and may favor facultative anaerobes capable of

using nitrate as an alternative electron acceptor. Such a shift could explain the significant depletion of nitrate in both planted and unplanted soils, and the accumulation of ammonium in some treatments due to inhibited nitrification.

Altogether, these processes suggest that the addition of P3HB, especially at concentrations of 1% and above, induces a cascade of microbial and biochemical changes that disrupt the N cycle. The imbalance between C and N availability leads to microbial dominance in nutrient acquisition, outcompeting plants for available N, and ultimately contributing to the inhibition of plant growth.

Therefore, the third hypothesis was confirmed. Furthermore, we can assume that some microbial groups also responded to changes in soil N availability.

Maize plant response to P3HB

The combined impact of P3HB-induced changes in soil characteristics, ranging from altered microbial community composition and reduced diversity to enhanced C mineralization, nitrification inhibition, and diminished inorganic N and oxygen availability (Fig. 8), resulted in a significant suppression of maize growth starting already at medium P3HB concentrations (≥ 1%, Fig. S8). While reduced NO_3^- availability appeared to be the main limiting factor, the role of impaired soil aeration, due to elevated microbial and root respiration, also seems to be critical. In line with this, both maize biomass and plant height markedly decreased at higher P3HB doses, despite recently observed increases in N and phosphorus concentrations in plant tissues [33], likely reflecting nutrient accumulation under stress rather than effective nutrient assimilation. In addition, the cited study also pointed to intensified nutrient competition between roots, rhizosphere microbes, and P3HB degraders. This competitive pressure was mirrored in reduced nutrient fluxes from soil to plant biomass, with N uptake decreasing sharply already at 1% P3HB, and nearly collapsing at 5–10%. Last, it was shown that the rhizosphere may also have promoted more efficient microbial incorporation of P3HB-derived C, but at the cost of increased nutrient demand, further intensifying plant–microbe competition.

These findings suggest a critical threshold around 1% P3HB, beyond which P3HB-induced disruptions in soil biochemical cycles, especially N cycling and soil aeration, begin to severely constrain plant development. This threshold is impacted by the soil texture, as lighter soil types can better support the aeration and affect the rate of P3HB biodegradation [85]. To elucidate the key mechanisms and their temporal dynamics, future studies should prioritize high-resolution monitoring during the early growth stages (2–3 weeks after emergence), as preliminary data indicate this period may be decisive for determining plant fate under P3HB exposure (Text S2).

Conclusions

The contamination of soil with P3HB microplastics induced significant changes in the composition and functions of microbial communities following enhanced *C* mineralization. Procaryotic α-diversity was not affected by P3HB, whereas fungal α-diversity declined starting at medium P3HB levels. The composition of prokaryotic and fungal communities shifted from a balanced microbiome, including oligotrophic and nitrifying groups, toward prevailing copiotrophic taxa, with a sharper shift in the eucaryotic community. This shift enhanced respiratory mineralization of P3HB-contained C and likely led to local O_2 depletion, promoting the abundance of anaerobes impacting N-mineralization. The decrease in nitrifying taxa limited the availability of mineral N in soil, particularly nitrates, due to the reduced oxidation of ammonia and possible anaerobic N respiration. Medium and high P3HB doses probably exceeded a critical threshold, triggering a transition in microbial community composition and dependent nutrient transformation from nutrient sequestration and availability to over-enhanced nutrient consumption, followed by the inhibition of plant growth.

A general recommendation can be derived for the safe limits of P3HB-based material use in agriculture. The critical effects started at approximately 1% of P3HB microplastics in soil. Therefore, for example, the application of 100 µm thick P3HB film, considering its decomposition and mixing with topsoil (10 cm), will result in P3HB content of about 0.1%, which corresponds to a safe dose. However, repeated applications of P3HB without allowing adequate time for its complete biodegradation between applications can increase its soil content to levels greater than or equal to 1%. Such accumulation may negatively impact soil quality and fertility over time.

Abbreviations

AGB	Aboveground biomass (dry weight)
AOB	Ammonia-oxidizing bacteria
Arg-IR	Soil respiration induced by L-arginine
BPs	Biodegradable plastics
CCA	Canonical correspondence analysis
Cit-IR	Soil respiration induced by citric acid
c(P3HB)	Soil content of poly-3-hydroxybutyrate
DM	Dry matter
NAG	Soil N-acetyl-β-D-glucosaminidase
NAG-IR	Soil respiration induced by N-acetyl-β-D-glucosamine
NH_4–N	Soil–ammonium–nitrogen
N_{min}	Soil inorganic nitrogen
NO_3–N	Soil–nitrate–nitrogen
P3HB	Poly-3-hydroxybutyrate
PCoA	Principal coordinate analysis
PHAs	Polyhydroxyalkanoates
phaZ	QPCR-detected microbial degraders of P3HB
PHBV	Poly(3-hydroxybutyrate-co-3-hydroxyvalerate)
Tre-IR	Soil respiration induced by D-trehalose

Supplementary Information

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Additional file 1.

Author contributions

Conceptualization: T.H., J.H., J.K. and M.B.; data curation: T.H., J.S.; formal analysis: T.B., A.K., J.S.; funding acquisition: M.B., J.K. and M.K.; investigation: T.H., J.H. and M.B.; methodology: T.H., J.H. and M.B.; project administration: M.B.; resources: J.H., T.H., J.S. and A.K.; software: T.B., T.H. and J.H.; supervision: V.P., J.K., M.K.; validation: V.P., J.K., M.K., A.K., M.B.; visualization: T.H., J.H., M.K. and V.P.; writing—original draft: J.H., M.B.; writing—review and editing: M.B., V.P., J.K., M.K.

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Data availability

The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

Declarations

Competing interests

The authors declare no competing interests.

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