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Nutrient recovery from African catfish sludge via aerobic mineralization and assessment of its fertilization potential in hydroponic basil cultivation

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ABSTRACT

The increasing demand for sustainable fertilizer sources highlights the potential of aquaculture by-products as alternatives to conventional mineral solutions in hydroponics. This study evaluated the fertilization potential of a liquid solution produced by aerobic digestion of fish sludge from a recirculating aquaculture system with African catfish (*Clarias gariepinus*) for indoor cultivation of basil (*Ocimum basilicum* L.) in a deep water culture system. Three nutrient solutions were compared: a conventional mineral solution (HYDRO), fish excrement mineralizate obtained by aerobic digestion (FEM), and a 1:1 (v/v) mixture of both (MIX). Cultivation with FEM alone resulted in a 17.5 % reduction in fresh shoot biomass and an overall reduction in N and P content in plant biomass compared to the hydroponic control, likely due to deficient potassium and iron levels in the FEM solution. However, plants grown in the FEM solution exhibited high potassium use efficiency and lower nitrate accumulation in plant tissues. The MIX variant achieved fresh shoot biomass comparable to the mineral control, enhanced root growth (+40.3 %), and reduced nitrate accumulation (−9.4 %) while maintaining vitamin C content and total antioxidant capacity. Fluorescence in situ hybridization confirmed the presence of *Bacillus* spp. and *Azospirillum* spp. in the FEM and MIX solutions, suggesting a potential biostimulant effect that needs further investigation. These findings indicate that FEM cannot serve as a full replacement for mineral nutrient solutions; however, appropriate supplementation with mineral fertilizers can achieve growth performance comparable to or exceeding that in mineral solutions, offering a sustainable and balanced cultivation strategy.

1. Introduction

The global demand for high-quality food is increasing steadily, while arable land is diminishing, freshwater availability is becoming more limited, and climate change poses growing threats to agriculture. In response, more effective crop production systems are gaining attention. Among them, hydroponics—a soilless cultivation method—offers advantages such as efficient water use, high productivity and suitability for cultivation in barren or urban environments [1]. Despite its advantages, hydroponic cultivation commonly uses nutrient solutions derived from mineral fertilizers, whose production is energy-intensive and frequently reliant on non-renewable mineral resources [2,3]. Moreover, the availability of these fertilizers is increasingly constrained by market volatility, geopolitical factors, and global crises [4,5]. Consequently, there is a growing need for sustainable alternatives that recover nutrients from organic waste streams. Most research to date has focused on livestock manure [6], [7], [8] or food processing by-products [9,10], with nutrients being released through aerobic or anaerobic digestion.

A promising but less explored source of nutrients is fish sludge, particularly within recirculating aquaculture systems (RAS). In aquaponics, RAS solution is directly used for plant cultivation, allowing for partial nutrient recovery. However, solid sludge—composed mainly of fish excrements and uneaten feed—is typically discarded [11]. Aquaponic systems also face challenges, including low nutrient concentrations [12,13], conflicting pH requirements for fish and plant parts of the system [14], and sodium accumulation [15]. Additionally, not all RAS operators can implement fully integrated systems. Importantly, the majority of nutrients—particularly nitrogen and phosphorus—are retained in the solid sludge, which has demonstrated potential as a fertilizer substrate [16]. Compared to manure from warm-blooded livestock, fish sludge contains a lower microbiological risk due to the absence of common pathogens such as *Escherichia coli* and *Salmonella* spp [17].

Several studies have investigated the mineralization of fish sludge by

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aerobic digestion (AD) [11, 18–24] though direct comparisons with conventional mineral solutions in plant cultivation remain limited. For instance, Lobanov et al. [25] reported lower lettuce yields but higher mineral content (Mg, Ca, Na, Si) when using digestates of fish sludge from RAS with rainbow trout (*Oncorhynchus mykiss*). Similarly, Ezziddine, Liltved & Seljåsen [26] observed a comparable yield but higher P, K, Ca, S, and B concentrations in lettuce grown on a solution derived from aerobically digested sludge originating from brown trout (*Salmo trutta*) rearing, relative to lettuce grown hydroponically.

Delaide et al. [27] demonstrated that a complemented aquaponic solution from a tilapia RAS, enriched with mineral nutrients, exhibited a 39 % faster growth rate than the control hydroponic variant and a 36 % higher root fresh weight. The improvement was attributed to the presence of plant growth-promoting microorganisms originating from the organic fraction of the solution. Furthermore, Khalil et al. [28] investigated the microbiota in water derived from tilapia and African catfish (*Clarias gariepinus*) rearing in a RAS and found that the solution was rich in bacterial and fungal microorganisms exhibiting antagonistic activity against several plant pathogens. Two microbial taxa frequently associated with enhanced plant growth are *Bacillus* and *Azospirillum* spp. These rhizobacteria can improve nutrient uptake and offer protection against pathogens [29,30]. Members of the genus *Bacillus* have been detected in the gut microbiome of African catfish [31], and its probiotic properties for this species have been confirmed [32]. While *Azospirillum* spp. are primarily soil bacteria, their presence has also been documented in RAS with African catfish [28].

Despite the growing production of African catfish in RAS [33,34], limited research has addressed the potential reuse of its fish sludge for hydroponic crop fertilization.To the best of our knowledge, this is the first study to evaluate the application of liquid fertilizer derived from aerobically digested fish sludge from African catfish RAS for the hydroponic cultivation of basil in an isolated system. Green basil (*Ocimum basilicum*) is a high-value herb of increasing commercial and scientific interest due to its bioactive compounds. Extracts of basil have demonstrated antimicrobial [35], insecticidal [36,37], and antifungal activity [38], making the plant relevant for food, pharmaceutical, and fragrance applications.

The objective of this study was to determine whether such organic-based fertilizer can serve as an effective alternative to conventional mineral solutions in deep water culture (DWC) basil production. We also investigated the presence of beneficial microorganisms (*Bacillus* spp. and *Azospirillum* spp.) on the root surface of basil, hypothesizing that their natural occurrence in the digestate may contribute to enhanced plant performance through biostimulant effects.

2. Materials and methods

2.1. Production of liquid fertilizer by aerobic digestion of fish sludge

Fish sludge originating from African catfish rearing in a RAS at the experimental facility of the Department of Fisheries and Hydrobiology (Mendel University in Brno, Zemědělská 1, 613 00 Brno, Czech Republic), was used as the organic substrate for liquid fertilizer production. The RAS consisted of three circular polypropylene tanks (1000 L each; total rearing volume 3000 L) connected to a biological filter unit (Nexus EAZY 320, Evolution Aqua Ltd, Wigan, UK). During the culture period, water temperature ranged from 25.4 to 26.1 °C, dissolved oxygen from 4.9 to 6.2 mg/L, and pH from 6.9 to 7.3. Stocking density was maintained at 230–240 kg/m^3. The fish used for sludge production were adult African catfish with an average body weight of approximately 1.0–1.2 kg, corresponding to approximately 200 fish per tank.

Fish were fed a commercial floating diet with a pellet size of 4.5 mm (Skretting ME-4.5 Meerval TOP, Nutreco N.V., Amersfoort, the Netherlands) at a rate of 2 % of body weight per day. According to the manufacturer, the feed consisted of processed poultry animal proteins, wheat, wheat gluten, soybean meal, fish meal, poultry fat, fish oil, and wheat starch. The declared nutrient composition was 44 % crude protein, 14 % crude fat, 1.4 % crude fibre, 9.5 % crude ash, 2.5 % calcium, 0.4 % sodium, and 1.2 % phosphorus. Most of the uneaten feed was manually removed on a daily basis to prevent its high abundance in the sludge. Consequently, the solid waste used in this study consisted predominantly of fish excrements. The effluent used in this study was collected at the site of the biological filter, where a vortex separator was installed for sedimentation and continuous collection of fish sludge. The sludge was dewatered by sedimentation and decantation. Excess water was subsequently removed by drainage through a 50 μm polypropylene textile to achieve approximately 10 % dry matter content, after which the material was stored at 4 °C until use in the experiment.

During waste collection and storage, a mineralization reactor was constructed (Fig. 1). The reactor was built from a 220 L polyethylene barrel (height 98 cm, Ø 49 cm). Two cylindrical air stones (300 × 50 mm, Ø 5 mm connection) were installed at the bottom and connected to a membrane compressor Jebao PA-35 (Jebao Co., Ltd., Zhongshan, China) to ensure continuous aeration and oxygen supply. To promote suspension mixing, an airlift made of polypropylene tubing (Ø 40 mm, 800 mm in length) was installed in the center of the reactor and stabilized with support pipes fixed to the upper part of the reactor wall. Air injected at the base of the airlift generated upward liquid flow, creating vertical circulation and homogeneous mixing without mechanical stirrers.

Before use, the refrigerated fish sludge was manually homogenized with a spatula to obtain a representative, uniform structure of the batch. The reactor was then loaded with 180 L of RAS water and 18 kg of dewatered fish sludge, containing 10 % dry matter, which corresponded to 1.8 kg of dry matter. Dry matter content was determined gravimetrically by drying representative samples at 105 °C to constant weight. Aerobic digestion was carried out for the following 21 days. During this period, the reactor contents were continuously aerated and mixed using an airlift, while maintaining the temperature at 30 °C with a 200 W heater equipped with a thermostat (OASE GmbH, Hörstel, Germany). No pH adjustment or microbial inoculum was applied during the aerobic digestion process. This procedure was chosen based on the work of

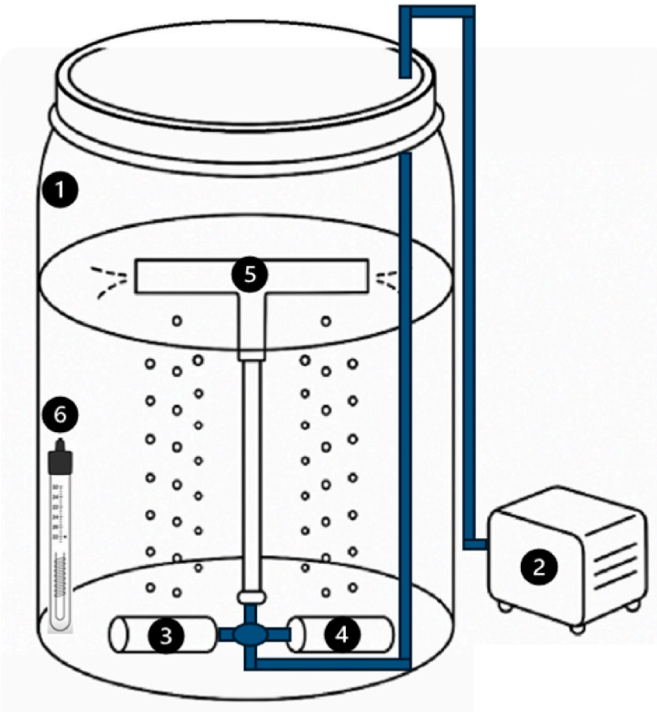

Fig. 1. Mineralization reactor design: (1) reactor vessel; (2) membrane air compressor; (3, 4) air stones; (5) airlift; (6) heater with thermostat.

Harabiš et al. [18], in which the aerobic digestion process was optimized.

After 21 days, aeration was stopped, the airlift removed, and the suspension was allowed to settle for 1 h. Subsequently, 80 L of supernatant was decanted into a secondary container and further clarified by gravity filtration through a 50 μm polypropylene textile to remove suspended fine particles. This process yielded a clear nutrient solution, which was then subjected to chemical analysis. The undiluted fish excrement mineralizate (UFEM) exhibited very high nutrient levels; therefore, prior to experimental use, UFEM was diluted 1:3 (v/v) with tap water, resulting in the FEM variant (see section 3.1).

2.2. Experimental system setup and cultivation conditions

The growth experiment was conducted in three identical indoor vertical hydroponic systems at the Department of Fisheries and Hydrobiology (Mendel University in Brno, Zemědělská 1, 61300 Brno, Czech Republic). One cultivation system consisted of three independent racks, each operating as a DWC unit. Every rack was connected to its own sump tank and a pump, forming a closed cultivation system with a total volume of 190 L (Fig. 2). The schematic of the cultivation system, consisting of three racks and thus three closed nutrient solution loops, is shown also in Fig. 2. Water in each system circulated continuously: a submersible pump delivered the solution to the respective rack and gravity returned it back to the sump tank. Each rack was equipped with LED lighting panels (red:blue ratio 2.5:1) positioned 220 mm above the plant canopy. The air temperature of the cultivation room was maintained at 20–23 °C using an air conditioning unit, and relative humidity was kept at 60–65 %. No supplemental CO_2 was provided.

2.3. Experimental design

The experiment was conducted in three identical vertical cultivation systems, each comprising three racks with three closed nutrient solution loops. Thus, the number of replicates for each nutrient solution variant was n = 3. Solution variants were rotated among the racks in each system to minimize positional effect. Basil seedlings were randomly assigned to the respective nutrient solution variants, with 20 plants per variant per replicate. Border plants were excluded from harvesting and evaluation; thus, for each variant and replicate, only six plants were used (18 plants per triplicate). Therefore, out of the total 180 planted plants, 54 plants were used for evaluation and analyses. Fig. 2 illustrates one of the arrangements of nutrient solution variants across the three racks within a single cultivation system. The three nutrient solution variants were as follows:

A. HYDRO (mineral control) – commercial hydroponic fertilizer for leafy vegetables, composed of two components: JUNGLE Garden G1

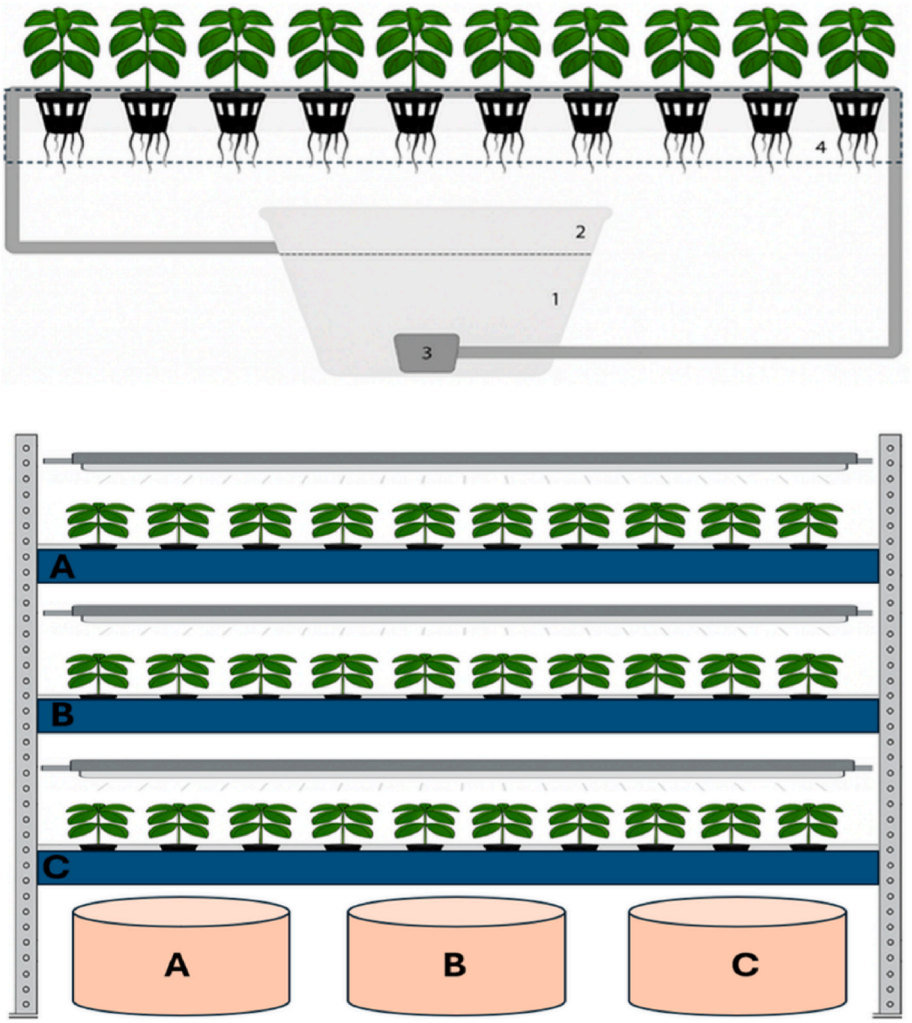

Fig. 2. Experimental setup and arrangement of nutrient solution variants within the cultivation system. Schematic of a single closed-loop nutrient solution circulation system showing: (1) sump tank; (2) nutrient solution level; (3) pump; and (4) cultivation rack. One of the three possible arrangements of nutrient solution variants among the three racks within a single cultivation system: (A) HYDRO; (B) FEM; (C) MIX.

(0.4 % N, 2 % P_2O_5, 4.5 % K_2O; Numazon Ltd., Brno, Czech Republic) and JUNGLE Garden BASE (7 % N, 11.2 % CaO, 0.22 % Fe; Numazon Ltd., Brno, Czech Republic). The solution was prepared by adding 2.5 mL JUNGLE Garden G1 and 1 mL JUNGLE Garden BASE per liter of tap water, as recommended by the manufacturer.

B. FEM (fish excrement mineralizate) – nutrient solution prepared from UFEM (undiluted fish excrement mineralizate) obtained by aerobic digestion of fish sludge from RAS with African catfish and diluted 1:3 (v/v) with tap water to obtain EC and nitrate values comparable to the HYDRO variant.

C. MIX – a 1:1 (v/v) mixture of HYDRO and FEM variant.

2.4. Course of the experiment

The experiment started on 1 May 2024 with the sowing of green basil into 40 × 40 × 40 mm rockwool cubes (ROCKWOOL B.V., Roermond, Netherlands) using seeds supplied by SUBA SEEDS COMPANY S.p.A., Longiano, Italy. Ten seeds were sown per cube and immediately irrigated with the mineral hydroponic solution (HYDRO) to standardize conditions before assignment to solution variants. At the cotyledon stage, the plants were thinned to five seedlings per cube. This standardization ensured uniform planting density and homogeneity across variants prior to transplanting into the cultivation systems. The seedlings were pre-grown for 21 days under standardized lighting conditions (18 h light/6 h dark, photosynthetic photon flux density ~ 130 μmol/m^2/s, with a red-to-blue light ratio of 2.5:1). The temperature in the cultivation room was maintained at 20–23 °C, with a relative humidity of 60–65 %. No supplemental CO_2 was provided.

Before transplanting the seedlings, all hydroponic systems were disinfected with 1 % (v/v) sodium hypochlorite and thoroughly rinsed with tap water to remove chlorine residues. On 22 May 2024, experimental cultivation systems (9 separate closed loops for nutrient solution, 190 L each) were filled with the respective nutrient solutions (see section 2.3).

Prior to transplanting the seedlings, the pH of all nutrient solutions (HYDRO, FEM, MIX) was adjusted from ~7.2 to 5.9 using 15 % (v/v) HNO_3 (Sigma-Aldrich Corporation, St. Louis, United States), which resulted in a slight increase in nitrate concentrations in all variants. The initial parameters of all nutrient solution variants before acidification are reported in Supplementary Table 1, while the values after acidification correspond to the T_0 values (see section 3.1). After adjustment, NO_3^- concentrations were comparable across variants (~560–575 mg/L).

Seedlings were transplanted into DWC systems and cultivated for 21 days under the same conditions as in the pre-growing phase. Throughout the experiment, pH, solution temperature and EC were measured daily. pH and nutrient solution temperature were measured using a HACH HQ40d multimeter (Hach Company Inc., Loveland, United States), while electrical conductivity (EC) was determined with a Hanna Combo HI98130 m (Hanna Instruments Inc., Smithfield, United States). Air temperature and relative humidity were also recorded daily. Evapotranspiration losses were compensated daily with deionized water to maintain constant system volume. At harvest (21 days after transplanting), deionized water was added for the last time to restore the original volume. The solution was then allowed to circulate within the systems to ensure proper mixing, and nutrient solution samples were collected, corresponding to T_1 values (see section 3.1).

2.5. Analytical methods

Detailed methodologies for the following sections are provided in the Supplementary Material.

2.5.1. Chemical analyses of solutions

Concentrations of NH_4^+–N, NO_2^-–N, NO_3^-–N, PO_4^{3-}–P, Cl^-, and dissolved Fe were determined spectrophotometrically according to APHA Standard Methods [39] using a PhotoLab 6600 UV–VIS spectrophotometer (WTW GmbH, Germany). Potassium (K^+), calcium (Ca^{2+}), and magnesium (Mg^{2+}) were measured colorimetrically using a Palintest Photometer 7100 (Halma Holdings Ltd., UK) following the manufacturer's protocols.

2.5.2. Photosynthetic, morphological, and compositional analyses of plants

Normalized Difference Vegetation Index (NDVI) and Quantum Yield (Qy) were measured before harvest using the handheld PlantPen NDVI 300 and FluorPen FP 110-LM/D devices (Photon Systems Instruments Ltd., Drásov, Czech Republic). All morphological parameters were measured using a balance (accuracy 0.01 g) or a ruler with a precision of 1 mm. The determination of total antioxidant capacity (TAC DPPH) was carried out according to the methodology of Zloch, Čelakovský & Aujezdská [40]. Vitamin C was analyzed using high-performance liquid chromatography (ECOM Ltd., Prague, Czech Republic) equipped with an S-2600 detector (KNAUER Scientific Instruments Ltd., Berlin, Germany) and an ARION® Polar C18 column (5 μm, 150 mm × 4.6 mm). The fiber content was determined according to the standard protocol provided by FibreBag. The determination of total nitrogen (N), phosphorus (P), and nitrates in plant samples was performed according to the methods described by Zbíral [41]. Total potassium (K) in the samples was determined by capillary isotachophoresis using an IONOSEP 2003 instrument (Recman–Laboratory Technique Ltd., Ostrava, Czech Republic) according to the manufacturer's application note. Plant pigments (chlorophyll *a*, chlorophyll *b*, and carotenoids) were determined spectrophotometrically according to Holm [42].

2.5.3. Calculation of nutrient use efficiency

The efficiency of nitrogen (NUE), phosphorus (PUE), and potassium (KUE) utilization was calculated as the percentage ratio between the nutrient accumulated in the plant and the nutrient supplied in the nutrient solution:

$$XUE\ (\%) = \frac{\text{nutrient accumulation in the plant [mg/plant]}}{\text{nutrient supplied [mg/plant]}} \times 100,$$

where *X* represents N, P, or K.

Nutrient accumulation was determined from the nutrient concentration in the dry shoot biomass (mg/kg DW, standardized to 100 % dry matter) multiplied by the mean dry shoot biomass per plant (g). The supplied amount was calculated as the difference in nutrient concentration between the start (T_0) and the end (T_1) of cultivation, multiplied by the system volume (190 L), and divided by the number of plants (n = 20). For nitrogen, all determined inorganic forms (NO_3^-, NH_4^+, NO_2^-) were included and converted to elemental N; phosphorus was calculated from PO_4^{3-} to elemental P; potassium was expressed directly as K^+.

2.5.4. Fluorescence in situ hybridization

Fluorescence in situ hybridization (FISH) was used to evaluate the presence of plant growth-promoting microorganisms (PGPMs) associated with plant roots. All samples and pure cultures were first hybridized with Bs-Cy3 and Bs-FAM probes (Eurofins Genomics, Brno, Czech Republic) according to Posada et al. [43], followed by hybridization with the AZO440a + probe labeled with Cy5 (Eurofins Genomics, Brno, Czech Republic) according to Stoffels, Castellanos & Hartmann [44]. Samples were analyzed using a confocal laser scanning microscope (LSM800, Carl Zeiss AG, Oberkochen, Germany). The abundance of *Bacillus* and *Azospirillum* spp. in basil root samples was evaluated on a 0–5 scale according to Alarcón & Cuenca [45].

2.6. Statistical analyses

Statistical analyses were performed in Statistica 14 software (DataBon Ltd., Prague, Czech Republic). Outliers were first removed from the dataset using a box plot with a coefficient of 1.5. One plant in the

HYDRO variant did not initially reach the nutrient solution after transplanting, resulting in reduced growth and extreme values across measured parameters, and was therefore excluded from the statistical analyses as an outlier. The remaining data were then tested for normality using the Shapiro-Wilk test ($\alpha = 0.05$) and for homogeneity of variance using Levene's test ($\alpha = 0.05$). Data that met the assumptions of normality and homogeneity were further analyzed using one-way analysis of variance (ANOVA). Statistically significant differences were detected using Tukey's HSD test ($\alpha = 0.05$). For data that did not meet one or both assumptions, non-parametric testing was performed using Kruskal–Wallis ANOVA, followed by a post-hoc multiple comparisons of mean ranks across all groups ($\alpha = 0.05$). Significant differences between groups in tables and figures are marked with lowercase (parametric) or uppercase (non-parametric) letters.

3. Results and discussion

3.1. Aerobic mineralization and characteristics of nutrient solutions

The mineralization reactor was fed with 180 L of RAS solution and 18 kg of dewatered sludge from the same African catfish system. In order to evaluate the enrichment of the original RAS solution with nutrients derived from solid sludge, the pH and EC values as well as the nutrient content of the RAS solution were analyzed prior to its introduction into the reactor (Table 1). After 21 days of aerobic digestion, the pH of the final solution decreased to a slightly acidic level (Table 1), which is consistent with ongoing nitrification. Nutrient concentrations increased substantially in the resulting UFEM, as reflected by the rise in EC from 2.05 mS/cm to 4.60 mS/cm. This increase was primarily driven by nitrates formed from organic nitrogen in the fish sludge, which was first converted to ammonia and subsequently oxidized to nitrates, representing a 266.7 % increase. In parallel, ammonium originally present in the RAS solution was almost entirely converted to nitrates, leaving only trace amounts in UFEM and indicating near-complete nitrification. Given the almost complete nitrification, it can be assumed that the mineralization reactor load contained a substantial proportion of heterotrophic bacteria, likely dominated by Proteobacteria, Bacteroidetes, and Firmicutes, as reported by Clols-Fuentes et al. [33]. The load also likely included ammonia- and nitrite-oxidizing bacteria (AOB and NOB), which carry out ammonia oxidation via the canonical two-step pathway, as well as representatives of comammox (Complete Ammonium Oxidation) microorganisms [46]. The presence of trace amounts of NH_4^+ in UFEM, however, also suggests that, alongside dominant nitrification, additional microbial processes may have occurred in the solution. In this context, the formation of localized oxygen-limited, or even anoxic, microenvironments within suspended sludge particles or microbial aggregates cannot be excluded, as oxygen stratification inside sludge flocs has been described even under aerated conditions [47]. Under such conditions, some facultatively anaerobic Bacillus spp. may remain metabolically active and may theoretically contribute to ammonium formation via dissimilatory nitrate reduction [48], while Azospirillum spp. may persist under microaerobic conditions, where nitrogen fixation can occur [49]. The occurrence of these taxa is further discussed in the FISH section.

Table 1
Comparison of the chemical composition of (1) the RAS solution originating from African catfish rearing, (2) the undiluted fish excrement mineralizate (UFEM) after 21 days of aerobic digestion in the reactor, and (3) the fish excrement mineralizate after dilution with tap water at a 1:3 (v/v) ratio (FEM) and before acidification.

Parameter	RAS solution	UFEM (before dilution)	FEM (after dilution, before acidification)
pH	7.05 (±0.01)	5.95 (±0.02)	7.2 (±0.02)
EC [mS/cm]	2.05 (±0.03)	4.60 (±0.04)	1.50 (±0.02)
NO_3^- [mg/L]	531.43 (±6.60)	1948.48 (±2.95)	487.12 (±0.57)
NH_4^+ [mg/L]	10.84 (±0.10)	1.84 (±0.08)	0.46 (±0.02)
PO_4^{3-} [mg/L]	237.85 (±1.90)	680.49 (±1.36)	170.12 (±0.36)
Cl^- [mg/L]	120.00 (±3.00)	234.63 (±1.20)	58.65 (±0.21)
K^+ [mg/L]	51.00 (±2.00)	86.48 (±1.80)	26.62 (±0.97)
Ca^{2+} [mg/L]	632.19 (±9.00)	940.94 (±3.11)	235.24 (±5.11)
Mg^{2+} [mg/L]	235.62 (±3.10)	243.22 (±2.51)	119.00 (±1.00)
Total dissolved Fe [mg/L]	0.040 (±0.020)	0.090 (±0.010)	0.09 (±0.03)

Note: Values are presented as mean ± SD (n = 3).

Aerobic digestion also released substantial amounts of phosphates (+186.1 %), chloride (+95.5 %), and calcium (+48.8 %) ions into UFEM. Notably, potassium ions and total iron—nutrients often limiting when RAS solutions are used for plant cultivation in aquaponic systems—were also released, increasing by 69.6 % and 125.0 %, respectively, compared to the original RAS solution. In contrast, magnesium was released only minimally, with an overall increase of just 3.2 %.

Because nitrate concentrations in UFEM substantially exceeded the recommended range for basil (≈443–664 mg/L NO_3^-; [50,51]), it was necessary to dilute it 1:3 (v/v) with tap water. This dilution also caused an increase in pH, which was therefore adjusted in all solutions prior to the experiment using 15 % (v/v) HNO_3 to a target value of 5.9, within the optimal range for hydroponics [52]. The dilution and acidification together produced FEM from UFEM (Tables 1 and 2) and ensured comparable nitrate levels, while the dilution further contributed to comparable phosphate and magnesium ion levels across all three solution variants (Table 2). Table 1 shows that after 21 days of aerobic digestion using the applied ratio of fish sludge to RAS water, a relatively concentrated solution (UFEM) was obtained. When comparing these results of our study with previously conducted similar studies, the solution obtained in our case was the most concentrated. In their first experiment, Delaide et al. [53] used sludge from pikeperch (*Sander lucioperca*) and obtained a significantly less concentrated solution through aerobic mineralization in terms of NO_3^- (−63.2 %), PO_4^{3-} (−98.5 %), K^+ (−87.3 %), Ca^{2+} (−68.7 %), and Mg^{2+} (−89.1 %); however, their solution contained higher levels of Cl^- (+19.1 %) and Fe (+65.6 %). Due to the lower concentrations, their resulting solution did not require further dilution for the lettuce growth test, nevertheless, it was concluded that the solution was deficient in terms of P, K, and Mn content. Similarly, the solution obtained by aerobic digestion of sludge from brown trout (*Salmo trutta*) farming in the study led by Ezziddine, Liltved & Seljåsen [26] generally contained substantially lower nutrient concentrations (except for K and Fe) and therefore did not require further dilution for the lettuce growth test. Aerobically mineralized sludge derived from Nile tilapia (*Oreochromis niloticus*) farming in the studies by Goddek et al. [54] and Tellbüscher et al. [55] was also characterized by low nutrient concentrations; the latter author explicitly highlights this limitation and emphasizes the need for future studies to focus on achieving higher degrees of remineralization. Although the solution obtained in our study was highly concentrated, its potential use in hydroponic systems is still associated with the transport of large volumes of solution when compared to highly concentrated two-component mineral liquid fertilizers. One approach to enhancing nutrient remineralization into the solution was demonstrated by Guyapale, Dionela & Huervana [21], who added an organic carbon source in the form of molasses to African catfish sludge in the mineralization reactor. As a result, after 15 days of aerobic mineralization, they obtained a solution containing 2700 mg N/L, 720 mg K/L, 12116 mg Ca/L, and 1696 mg Mg/L. Assuming that all detected nitrogen was in the form of nitrates, the final solution contained 514 % more NO_3^-, 733 % more K, 1188 % more Ca, and 597 % more Mg compared with our study. On the other hand, lower phosphorus mineralization was achieved in their study. Despite the often insufficient nutrient concentrations mentioned above, all of these results demonstrate that fish sludge represents a significant reservoir of nutrients that can be effectively recovered and reused in plant production.

Based on the aforementioned studies, it can be concluded that the

Table 2
Parameters of the nutrient solution variants measured at the beginning (T_0, after acidification) and at the end (T_1) of the experiment.

Nutrient solution	HYDRO		FEM		MIX	
Time	T_0	T_1	T_0	T_1	T_0	T_1
pH	5.90 (±0.01)	6.18 (±0.02)	5.90 (±0.02)	6.31 (±0.02)	5.90 (±0.02)	6.26 (±0.02)
EC [mS/cm]	1.78 (±0.03)	1.50 (±0.03)	1.60 (±0.03)	1.38 (±0.02)	1.71 (±0.03)	1.44 (±0.02)
NO_3^- [mg/L]	572.27 (±10.40)	366.96 (±0.25)	563.02 (±0.57)	349.52 (±0.42)	575.12 (±0.67)	345.08 (±0.21)
NH_4^+ [mg/L]	5.44 (±0.07)	0.78 (±0.04)	0.46 (±0.02)	0.12 (±0.02)	3.15 (±0.06)	0.39 (±0.02)
PO_4^{3-} [mg/L]	182.04 (±0.77)	134.59 (±0.09)	170.12 (±0.36)	123.28 (±0.25)	170.81 (±0.29)	121.91 (±0.22)
Cl^- [mg/L]	26.94 (±0.10)	16.63 (±0.04)	58.65 (±0.21)	48.35 (±0.18)	45.83 (±0.20)	36.42 (±0.17)
K^+ [mg/L]	115.00 (±1.00)	55.00 (±1.00)	26.62 (±0.97)	5.04 (±0.12)	60.33 (±0.58)	23.00 (±1.00)
Ca^{2+} [mg/L]	111.33 (±0.58)	34.00 (±1.00)	235.24 (±5.11)	192.39 (±1.93)	228.83 (±15.05)	157.56 (±13.75)
Mg^{2+} [mg/L]	117.88 (±2.65)	95.88 (±11.07)	119.00 (±1.00)	90.80 (±0.72)	124.83 (±3.32)	87.42 (±34.41)
Total dissolved Fe [mg/L]	2.20 (±0.22)	0.40 (±0.04)	0.09 (±0.03)	0.04 (±0.01)	1.10 (±0.17)	0.20 (±0.03)

Note: Values are presented as mean ± SD (n = 3).

quality of the final product of aerobic mineralization varies considerably depending on:

(1) the aerobic mineralization technology, including: (a) the ratio of sludge dry matter to liquid; (b) the source of the liquid (tap water vs. RAS solution); (c) aerobic digestion conditions, such as temperature, dissolved oxygen, pH and its adjustment, the addition of activated sludge or organic carbon sources, and digestion time;
(2) the RAS production technology, including: (a) fish species and age; (b) stocking density, feed type and feeding regime; (c) RAS system configuration, including flow regulation, sludge collection and treatment methods [26].

Parameters of the three nutrient solution variants at the start (T_0) and end (T_1) of basil cultivation are summarized in Table 2. During cultivation, pH increased slightly, reaching 6.18–6.31 at T_1. This rise can be attributed to nitrate assimilation by plants, where anion uptake leads to OH^- and HCO_3^- release [56], and to microbial activity in FEM and MIX, where NO_3^- reduction increases alkalinity [57,58]. Nevertheless, pH remained within the optimal range of 5.5–6.5 for hydroponic basil [52]. The ability to adjust pH to this level represents a distinct advantage of using aerobically digested sludge, whereas in coupled aquaponics, pH must remain near neutral as a compromise between fish, nitrifying bacteria and plants [13].

Initial EC values ranged from 1.60 mS/cm (FEM) to 1.78 mS/cm (HYDRO), with MIX intermediate at 1.71 mS/cm. EC gradually decreased in all variants, with the greatest relative reduction observed in HYDRO (−16 %), reflecting intensive uptake of ions. In all variants, EC remained within the optimal range for basil hydroponics (1.3–2.0 mS/cm; [59,60]). However, EC alone does not ensure a balanced ionic composition. Differences among the variants were therefore more strongly driven by their specific ionic profiles: HYDRO showed the largest decline because of its balanced composition and optimal nutrient availability, whereas FEM — characterized by low K^+ and Fe — exhibited a smaller relative change. In FEM, K^+ and Fe were likely the limiting ions due to their reduced concentrations resulting from the applied dilution; once they were depleted, plant metabolism and growth slowed, thereby reducing the uptake of other nutrients as well.

FEM contained sufficient NO_3^- and PO_4^{3-}, consistent with findings of Delaide et al. [19] and Harabiš et al. [18], who reported that these macronutrients are predominantly released into the aqueous phase during aerobic sludge digestion. The elevated Cl^- concentration in FEM resulted from NaCl addition in the RAS, where it is routinely applied to prevent infections and support fish osmoregulation, with recommended levels above 100 mg/L [61]. For plants, Cl^- has an ambivalent role: it is an essential micronutrient and cofactor of photosystem II, but in high concentrations can compete with NO_3^- and SO_4^{2-} uptake and cause ion stress [62]. Nevertheless, it was observed that basil can tolerate levels higher than 2000 mg/L [63]. In this experiment, maximum Cl^- concentration in FEM was ~59 mg/L, more than 30-fold below toxicity thresholds. Thus, chloride likely did not limit basil growth and may have contributed to partial regulation of stomatal conductance and photosynthesis. The Ca:Mg:K ratios were 2:1:0.22 in FEM and 1.8:1:0.48 in MIX, compared to the recommended 4:1:4 for leafy vegetables [64]. The extremely high Ca:K ratios in FEM (≈9:1) and MIX (≈4:1), compared to the recommended 1:1, may have induced cationic antagonism, limiting K^+ and, to a lesser extent, Mg^{2+} uptake despite adequate Mg concentrations. Although Fe concentrations increased substantially in UFEM relative to the original RAS solution, the final FEM solution remained highly deficient in iron. This likely reflects generally low Fe levels in fish feed and, consequently, in fish sludge [16]. Accordingly, Fe concentrations in FEM were well below the recommended range for leafy vegetables (1–2 mg Fe/L), being approximately 10–20 times lower [65,66]. In contrast, this limitation was compensated for in the MIX variant.

Overall, all nutrient solutions maintained pH and EC within ranges suitable for hydroponic basil. FEM displayed nutritional imbalances (low K^+ and Fe, high Cl^-), while MIX partly corrected these deviations. These differences in ionic profile represented a key factor reflected in subsequent photosynthetic, morphological and compositional plant parameters.

3.2. Photosynthetic and morphological parameters

The measured photosynthetic parameters (Table 3) did not meet the assumptions required for ANOVA and were therefore evaluated using non-parametric tests. For the NDVI parameter, significant differences were observed between the HYDRO and FEM variants, with MIX representing an intermediate variant. The lower NDVI values observed in the organic-based variants may be attributed to unbalanced plant

Table 3
Photosynthetic and morphological parameters of basil grown under three nutrient solution variants: HYDRO, FEM and MIX.

Parameter	HYDRO	FEM	MIX
NDVI	0.68 ± 0.02 A	0.65 ± 0.04 B	0.67 ± 0.02 AB
Qy	0.82 ± 0.01 B	0.83 ± 0.01 B	0.84 ± 0.01 A
Fresh shoot biomass [g]	75.03 ± 8.12 a	61.90 ± 6.35 b	78.55 ± 7.33 a
Fresh stem biomass [g]	20.99 ± 2.93 ab	17.49 ± 3.46 b	23.11 ± 2.63 a
Fresh leaf biomass [g]	49.77 ± 5.24 a	40.23 ± 5.21 b	51.32 ± 3.44 a
Shoot length [cm]	31.28 ± 1.59 A	26.36 ± 1.11 B	31.14 ± 1.92 A
Root length [cm]	52.75 ± 6.04 b	38.97 ± 2.84 c	62.14 ± 5.69 a
Dry shoot biomass [%]	6.91 ± 0.29 a	7.22 ± 0.35 a	7.00 ± 0.29 a
Dry root biomass [g]	0.72 ± 0.11 c	1.51 ± 0.14 a	1.01 ± 0.11 b
Root:shoot ratio	0.13 ± 0.02 b	0.33 ± 0.06 a	0.17 ± 0.03 b

Note: Values are presented as mean ± SD (n = 18 for all parameters except fresh stem and leaf biomass, dry shoot biomass and root:shoot ratio, where n = 9), with outliers excluded from the analysis. Different lowercase letters in rows indicate statistically significant differences among variants acc. to parametric post-hoc Tukey's HSD test (α = 0.05), different uppercase letters in rows indicate statistically significant differences among variants acc. to non-parametric post-hoc multiple comparison of mean ranks (α = 0.05).

nutrition, including antagonistic effects of Ca^{2+} on K^{+} and Mg^{2+} uptake and Fe deficiency. These factors generally resulted in slightly (and not always significantly) reduced formation of photosynthetic pigments and, consequently, slightly lower NDVI values. Nevertheless, the differences were minor, and it can be concluded that plants across all variants exhibited high vigor and photosynthetic activity. Similarly, values of the quantum yield of Photosystem II, which reflects the efficiency of light utilization and serves as a sensitive stress indicator, were very similar across variants, with the highest values observed in the MIX variant. Comparable quantum yield results for basil grown under hydroponic and aquaponic conditions were reported by Signorini et al. [67].

Basil plants grown in the FEM solution exhibited the lowest fresh shoot biomass, being 17.5 % lower than in HYDRO and 21.2 % lower than in MIX (Table 3, Fig. 3). Comparable results were reported by Ezziddine, Liltved & Seljåsen [26] and Delaide et al. [53], where lettuce grown in organic solutions based on aerobically mineralized sludge exhibited 16 % and 22 % reductions in edible biomass, respectively, compared to the hydroponic control. In contrast, the MIX variant showed a higher mean biomass but did not differ significantly from the HYDRO variant, suggesting that mineral supplementation may represent an effective strategy to enhance the overall fertilization potential of the FEM solution. This result is supported by Tellbüscher et al. [55], who observed comparable fresh leaf biomass in lettuce grown in a mineral-supplemented solution based on aerobic digestate of Nile tilapia sludge compared to a hydroponic control.

The reduced fresh shoot biomass, as well as fresh stem and leaf biomass separately and shoot length, observed in FEM was likely caused by dilution of UFEM to FEM, which resulted in the limited availability of K^{+} and Fe in this variant. Potassium is essential for basil growth, its deficiency reduces biomass and shifts allocation towards root development [68]. This trend was confirmed by our study: the root dry biomass of the FEM variant was 109.7 % higher compared to the hydroponic control, whereas the root dry biomass of the MIX variant was 40.3 % higher than in the HYDRO variant. Consistentely, FEM exhibited the highest root:shoot ratio (0.33 ± 0.06) compared to MIX (0.17 ± 0.03) and HYDRO (0.13 ± 0.02). On the other hand, the FEM variant exhibited the shortest roots (Fig. 4). In combination with the higher root biomass, this may indicate alterations in root system architecture; however, as root volume, the number of root tips, and individual root diameter were not measured, it is difficult to determine whether the higher root dry mass observed in the FEM treatment (compared to the HYDRO treatment) resulted from more pronounced root branching, greater root thickness, or a combination of both. Considering the monitored solution parameters, the altered root morphology may have been influenced by the markedly different Ca:K ratio, which at the beginning of the experiment was 9:1 in the FEM treatment compared with 1:1 in the HYDRO treatment. Calcium is known to promote root-hair formation by regulating a Ca^{2+}–ROS signaling hub that coordinates cell-wall remodeling and cytoskeletal dynamics, thereby enabling controlled tip growth and strengthening of the cell wall [69]. In contrast, potassium deficiency can modify root system architecture by altering the transcription of K^{+} transporters, which disrupts auxin gradients in the root and triggers auxin redistribution that promotes lateral root formation as an adaptive response [70]. It can be hypothesized that the FEM solution may also contain additional bioactive compounds (e.g., phytohormones) that contributed to the observed effects on root morphology, which warrants further investigation.

3.3. Compositional parameters

Compositional analysis showed that total antioxidant capacity (DPPH), vitamin C, fiber, total K, and chlorophyll *b* in plant tissues did not differ significantly among the variants ($p \geq 0.05$; Table 4). From this perspective, basil grown in the organic-based solutions was of comparable quality to the hydroponic control. However, the organic variants

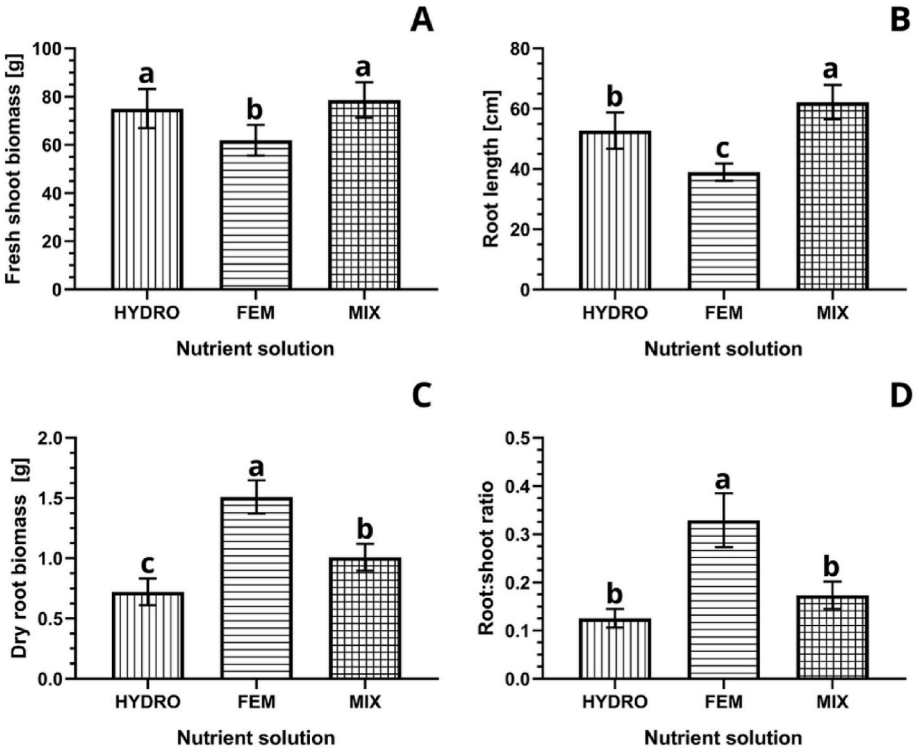

Fig. 3. Selected morphological parameters of basil grown under three nutrient solution variants: HYDRO, FEM and MIX: (A) fresh shoot biomass; (B) root lenght; (C) dry root biomass; and (D) root:shoot ratio.
Note: Values are presented as mean ± SD (for A, B and C, n = 18; for D, n = 9), with outliers excluded from the analysis. Different lowercase letters above columns indicate statistically significant differences among variants acc. to parametric post-hoc Tukey's HSD test (α = 0.05).

Fig. 4. Morphology of basil roots under three nutrient solution variants (HYDRO, MIX and FEM) after 21 days of cultivation in DWC system.

exhibited statistically significantly lower values for total N, P, chlorophyll *a*, and carotenoids. Conversely, in terms of nitrate content, basil grown in the organic-based solutions performed better (Table 4, Fig. 5).

Despite standardized nitrate concentrations at the start of the experiment, reduced nitrogen content was observed in FEM and MIX compared to HYDRO at the end. This likely resulted from imbalanced nutrition, particularly limited potassium, which can indirectly restrict nitrate uptake and translocation through coordinated K^+–NO_3^- transport regulation [71]. Our findings contrast with Ezziddine, Liltved & Seljåsen [26], who reported higher nitrogen content in lettuce grown on an organic solution (aerobically digested fish sludge) than in a hydroponic control, likely because none of the nutrients were limiting in their study. Although phosphate levels in the initial nutrient solutions were comparable, FEM plants showed significantly lower tissue P than the other variants. This reduction may be attributed to phosphate uptake by microorganisms or precipitation with the high calcium content in this variant, forming insoluble calcium phosphate ($Ca_3(PO_4)_2$) or dicalcium phosphate ($CaHPO_4$) [72]. Additionally, the low potassium and iron availability in FEM could have further limited phosphate uptake and transport, as K^+ is important for ion translocation and Fe supports root energy metabolism necessary for active nutrient acquisition. The content of photosynthetic pigments followed a similar pattern, with the highest levels in HYDRO and lower levels in the organic-based variants (chlorophyll *b* showed no significant difference). The differences were likely due to limited Fe availability in FEM and MIX, as iron is essential for chlorophyll biosynthesis and PSII function [68]. Similar effects of Fe deficiency on chlorophyll synthesis and biomass were reported by Kasozi et al. [58] in aquaponic systems. Nitrate concentrations in plant tissues reflect the balance between nitrate uptake and assimilation. In our study, nitrate accumulation closely followed potassium availability: FEM, with the lowest K (26.62 mg/L), had the lowest nitrate, MIX (60.33 mg/L K) was intermediate, and HYDRO (115.00 mg/L K) the highest. This pattern suggests that potassium limitation primarily restricted nitrate uptake and vacuolar storage [71], while lower total nitrogen in organic-based variants indicates that enhanced nitrate assimilation was not the main factor driving reduced nitrate levels in FEM and MIX. Lower NO_3^- accumulation in FEM and MIX represents a positive aspect of product quality.

3.4. Nutrient use efficiency

Differences in mineral composition were also reflected in nutrient use efficiency indices. FEM showed the lowest NUE (35.27 %) and PUE (6.80 %), while HYDRO achieved the highest values in case of NUE (45.87 %) and MIX in the case of PUE (9.21 %) (Table 5). Conversely, K^+ use efficiency (KUE) was highest in FEM (85.94 %). Although absolute K content in tissues was low, the limiting supply led to its maximal utilization. This adaptive mechanism corresponds to results in basil described by Mourantian et al. [73] and Attia et al. [68], where reduced biomass under low K supply was accompanied by increased relative uptake efficiency. Similarly, Patloková & Pokluda [74] reported that more than 70 % of supplied K was incorporated in aquaponic basil, substantially higher than in mineral variants. Goddek & Vermeulen [61] likewise observed stimulated K uptake in RAS-derived waters.

3.5. Fluorescence in situ hybridization

For the FISH staining, two probe sets were used:

Table 4
Compositional parameters of fresh basil grown under three nutrient solution variants: HYDRO, FEM and MIX.

Parameter	HYDRO	FEM	MIX
TAC DPPH [mg/kg]	775.28 ± 146.30 a	730.76 ± 130.28 a	675.03 ± 159.64 a
Vitamin C [mg/kg]	18.79 ± 5.99 a	16.90 ± 5.14 a	17.51 ± 4.42 a
Fiber [%]	1.73 ± 0.15 a	1.70 ± 0.20 a	1.54 ± 0.17 a
N [mg/kg]	2902.01 ± 197.63 a	2662.87 ± 166.42 b	2634.22 ± 114.14 b
P [mg/kg]	176.02 ± 11.45 a	161.93 ± 10.71 b	177.02 ± 13.49 a
K [mg/kg]	3116.39 ± 419.70 A	2887.02 ± 298.66 A	2983.71 ± 199.08 A
Chlorophyll *a* [mg/kg]	267.34 ± 13.44 A	248.34 ± 21.11 AB	225.42 ± 30.40 B
Chlorophyll *b* [mg/kg]	63.34 ± 6.46 a	55.47 ± 9.32 a	62.29 ± 8.63 a
Carotenoids [mg/kg]	119.38 ± 4.85 A	108.25 ± 10.48 AB	93.63 ± 11.19 B
Nitrates [mg/kg]	5464.32 ± 106.38 a	3848.12 ± 257.71 c	4950.86 ± 336.41 b

Note: Values are presented as mean ± SD (n = 9), with outliers excluded from the analysis. Different lowercase letters in rows indicate statistically significant differences among variants acc. to parametric post-hoc Tukey's HSD test (α = 0.05), different uppercase letters in rows indicate statistically significant differences among variants acc. to non-parametric post-hoc multiple comparison of mean ranks (α = 0.05).

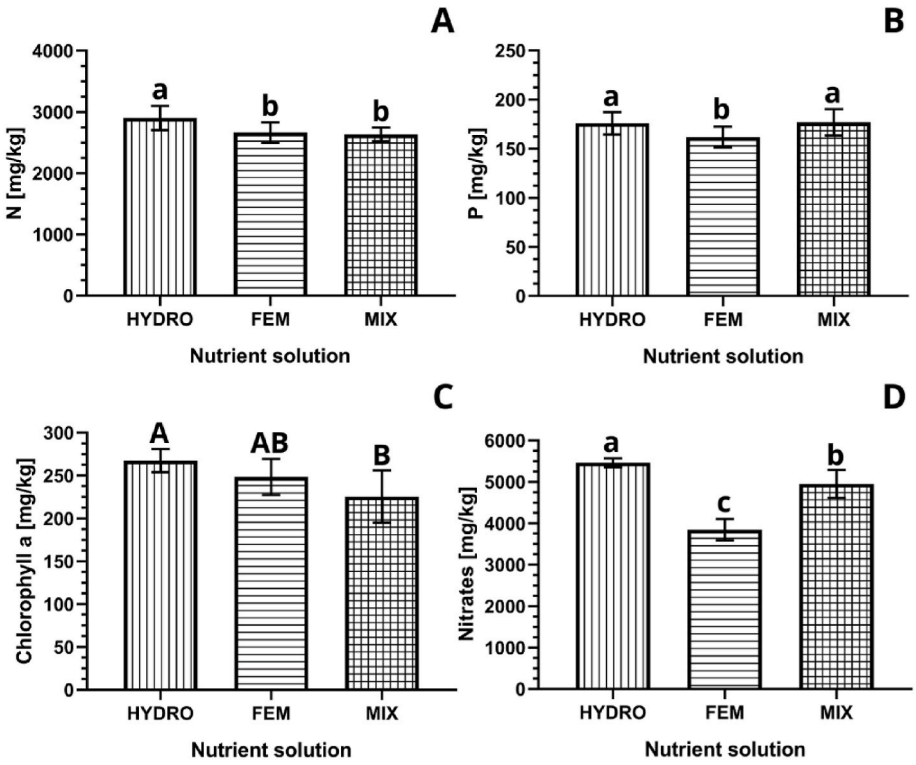

Fig. 5. Selected compositional parameters of basil grown under three nutrient solution variants: HYDRO, FEM and MIX: (A) nitrogen; (B) phosphorus; (C) chlorophyll *a*; and (D) nitrate content.
Note: Values are presented as mean ± SD (n = 9), with outliers excluded from the analysis. Different lowercase letters above columns indicate statistically significant differences among variants acc. to parametric post-hoc Tukey's HSD test (α = 0.05), different uppercase letters above columns indicate statistically significant differences among variants acc. to non-parametric post-hoc multiple comparison of mean ranks (α = 0.05).

Table 5
Nitrogen, phosphorus and potassium use efficiency (NUE, PUE, KUE; %) of basil grown under three nutrient solution variants: HYDRO, FEM and MIX.

Parameter	HYDRO	FEM	MIX
NUE (%)	45. 87 ± 6.08 a	35.27 ± 4.82 b	40.47 ± 3.86 ab
PUE (%)	9.02 ± 1.43 a	6.80 ± 0.90 b	9.21 ± 0.97 a
KUE (%)	40.97 ± 7.07 c	85.94 ± 13.55 a	66.48 ± 7.92 b

Note: Values are presented as mean ± SD (n = 9), with outliers excluded from the analysis. Different lowercase letters in rows indicate statistically significant differences among variants acc. to parametric post-hoc Tukey's HSD test (α = 0.05).

Table 6
Colonization of basil roots by *Bacillus* spp. and *Azospirillum* spp. (scale 0–5 according to Alarcón and Cuenca, [45]).

Variants	*Bacillus* spp.	*Azospirillum* spp.
HYDRO	0.0 ± 0.0	0.0 ± 0.0
FEM	2.8 ± 0.4	2.4 ± 0.5
MIX	0.4 ± 0.5	0.2 ± 0.4

Note: Values are presented as mean ± SD (n = 10).

1. Bs-Cy3 and Bs-FAM, designed by Posada et al. [43] for *Bacillus subtilis*;
2. the AZO440a + probe, designed by Stoffels, Castellanos & Hartmann [44] for the Azospirillum-Skermanella-Rhodocista cluster.

When the sequences of the first probe set are submitted to the NCBI Basic Local Alignment Search Tool [75], the results show that these probes also hybridize with other *Bacillus* species (e.g., *B. licheniformis*, *B. paralicheniformis*, *B. tequilensis*, *B. mojavensis*, etc.) as well as with several other genera, such as *Anaerobacillus*, *Gracilibacillus*, and *Brevibacillus*. The AZO440a + probe hybridizes with species such as *Azospirillum lipoferum*, *A. thermophilum*, *A. brasilense*, *A. formosense*, and others, and also with representatives of the genera *Skermanella*, *Arenibaculum*, and *Niveispirillum*. Thus, the detected bacteria may correspond to some of the species listed above; however, for simplicity, we refer to them here as *Bacillus* spp. and *Azospirillum* spp.

FISH analysis revealed differences in basil root colonization by *Bacillus* spp. and *Azospirillum* spp. among variants (Table 6, Fig. 6). No colonization was detected in the mineral control (HYDRO). In FEM, the presence of both bacterial groups was evident: *Bacillus* spp. reached a mean score of 2.8 ± 0.4 (corresponding to mean colonization of 42 %), while *Azospirillum* spp. averaged 2.4 ± 0.5 (corresponding to mean colonization of 26 %). In the MIX variant, colonization was recorded only sporadically (*Bacillus* spp. 0.4 ± 0.5; *Azospirillum* spp. 0.2 ± 0.4).

These results demonstrate that aerobically mineralized digestate (FEM) serves not only as a source of nutrients but also as a carrier of microbial communities, including *Bacillus* spp. and *Azospirillum* spp. The origin of these taxa in FEM and MIX can be unequivocally attributed to fish sludge, their occurrence in African catfish excrements has been confirmed previously [28–29,31]. The low intensity of colonization in MIX is likely related to the lower inoculation potential due to dilution of the organic fraction of FEM. These genera are known to produce phytohormones (auxins, cytokinins, gibberellins) and enhance nutrient uptake, which may explain the superior root development and photosynthetic performance observed in MIX, even under lower nutrient concentrations [29–30,76]. Similar synergistic effects of combined mineral–organic fertilization have been reported in basil and lettuce [27,77].

It should be noted that only two PGPM taxa were monitored, whereas digestate likely contains a broader spectrum of bacteria and fungi [33, 78] that may significantly influence basil growth. Moreover, the method

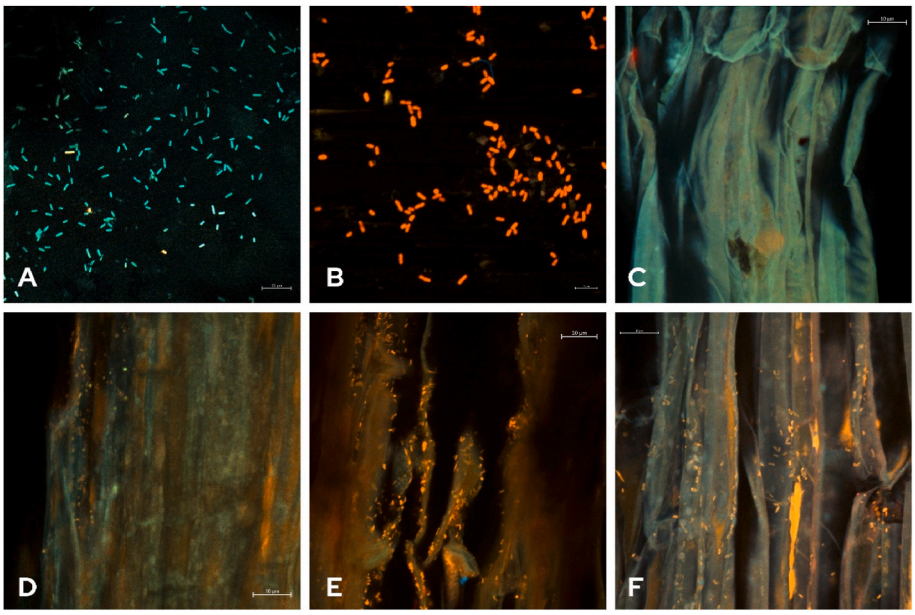

Fig. 6. Detection of *Bacillus* spp. and *Azospirillum* spp. in basil roots using FISH: (A) *Bacillus subtilis* – positive control; (B) *Azospirillum brasilense* – positive control; (C) negative, unstained control; (D) basil roots – HYDRO variant; (E) basil roots – FEM variant; (F) basil roots – MIX variant.

captured only microorganisms attached to root tissues, not those present in the nutrient solution, which may have behaved differently during cultivation. Consequently, it cannot be excluded that part of the microbial community in FEM and MIX remained in the aqueous phase or was flushed out during the trial. Future studies should combine FISH with molecular approaches (qPCR, NGS) targeting both the rhizosphere and the nutrient solution to better understand PGPM abundance in DWC systems.

3.6. Limitations of the study

The main limitation of this study is the need to dilute the original UFEM to obtain the resulting FEM, primarily due to the high nitrate concentration. Such elevated nitrate levels could potentially lead to plant damage and excessive nitrate accumulation in basil shoots. However, this dilution simultaneously reduced the concentrations of other nutrients, resulting in suboptimal levels of K^+ and total Fe in the final solution. Given that mineral nutrition in hydroponic herb production has been well optimized in previous studies, it is therefore advisable, when applying FEM-based solutions, to maintain recommended K and Fe concentrations and supplement these nutrients with mineral fertilizers if necessary. In addition to these compositional constraints, the interpretation of plant responses is limited by the lack of detailed root morphology measurements. Future studies should therefore further characterize root morphology to elucidate plant responses to aerobically mineralized fish sludge, including the effects of its complex nutrient profile and bioactive compounds. Finally, another limitation of the study is the relatively narrow scope of microbial analysis, as only two groups of PGPMs (*Bacillus* spp. and *Azospirillum* spp.) were monitored using FISH. Future research should thus include a broader microbial assessment to better characterize the microbial community developing during the aerobic digestion of fish sludge.

4. Conclusion

While the fish excrement mineralizate alone was insufficient to fully replace conventional mineral fertilization due to nutrient imbalances, its combination with a mineral nutrient solution enabled plant growth and yield comparable to the hydroponic control. The mixed fertilization strategy additionally promoted root development and reduced nitrate accumulation without compromising nutritional quality. However, further optimization of the mineralizate is required, particularly with respect to increasing nutrient concentration to improve the economic feasibility of storage and transport. Furthermore, the microbial community associated with the digestate-derived solution may contribute to biostimulatory effects; however, this hypothesis requires systematic experimental verification. Overall, the integration of aquaculture-derived mineralizates with targeted mineral nutrient supplementation offers a viable approach to improving nutrient circularity and sustainability in hydroponic systems.

CRediT authorship contribution statement

Lukáš Harabiš: Writing – original draft, Methodology, Funding acquisition, Data curation, Conceptualization. **Kateřina Patloková:** Writing – original draft, Validation, Methodology, Formal analysis, Data curation, Conceptualization. **Jan Mareš:** Supervision. **Robert Pokluda:** Validation, Supervision.

Ethics statement

This study did not involve experimental procedures on live fish. The fish sludge used in the experiment was obtained as a by-product of routine operation of a recirculating aquaculture system.

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.jafr.2026.102888.

Data availability

Data will be made available on request.

References

[1] H.M. Resh, Hydroponic Food Production: a Definitive Guidebook for the Advanced Home Gardener and the Commercial Hydroponic Grower, eighth ed., CRC Press, 2022.

[2] K. Chojnacka, K. Moustakas, A. Witek-Krowiak, Bio-based fertilizers: a practical approach towards circular economy, Bioresour. Technol. 295 (2020) 122223, https://doi.org/10.1016/j.biortech.2019.122223.

[3] I. Sigurnjak, E. Michels, S. Crappé, S. Buysens, F.M.G. Tack, E. Meers, Utilization of derivatives from nutrient recovery processes as alternatives for fossil-based mineral fertilizers in commercial greenhouse production of *Lactuca sativa* L, Sci. Hortic. 198 (2016) 267–276, https://doi.org/10.1016/j.scienta.2015.11.038.

[4] J. Peñuelas, F. Coello, J. Sardans, A better use of fertilizers is needed for global food security and environmental sustainability, Agric. Food Secur. 12 (2023) 5, https://doi.org/10.1186/s40066-023-00409-5.

[5] R. Bulgari, A. Petrini, G. Cocetta, C. Nicoletto, A. Ertani, P. Sambo, A. Ferrante, S. Nicola, The impact of COVID-19 on horticulture: critical issues and opportunities derived from an unexpected occurrence, Horticulturae 7 (6) (2021) 124, https://doi.org/10.3390/horticulturae7060124.

[6] Z. Liu, J. Howe, X. Wang, X. Liang, T. Runge, Use of dry dairy manure pellets as nutrient source for tomato (*Solanum lycopersicum* var. *cerasiforme*) growth in soilless media, Sustainability 11 (3) (2019) 811, https://doi.org/10.3390/su11030811.

[7] I. Szekely, Z. Zeaiter, M.H. Jijakli, Development of a simple bioponic method using manure and offering comparable lettuce yield than hydroponics, Water 15 (13) (2023) 2335, https://doi.org/10.3390/w15132335.

[8] P. Tikasz, S. MacPherson, V. Adamchuk, M. Lefsrud, Aerated chicken, cow, and Turkey manure extracts differentially affect lettuce and kale yield in hydroponics, Int. J. Recycl. Org. Waste Agric. 8 (3) (2019) 241–252, https://doi.org/10.1007/s40093-019-0261-y.

[9] C. Kawamura-Aoyama, K. Fujiwara, M. Shinohara, M. Takano, Study on the hydroponic culture of lettuce with microbially degraded solid food waste as a nitrate source, Jpn. Agric. Res. Q. 48 (1) (2014) 71–76, https://doi.org/10.6090/jarq.48.71.

[10] S. Wongkiew, C. Polprasert, P.L. Noophan, T. Koottatep, V. Kanokkantapong, K. C. Surendra, S.K. Khanal, Effects of vermicompost leachate on nitrogen, phosphorus, and microbiome in a food waste bioponic system, J. Environ. Manag. 339 (2023) 117860, https://doi.org/10.1016/j.jenvman.2023.117860.

[11] Z. Khiari, S. Kaluthota, N. Savidov, Aerobic bioconversion of aquaculture solid waste into liquid fertilizer: effects of bioprocess parameters on kinetics of nitrogen mineralization, Aquaculture 500 (2019) 492–499, https://doi.org/10.1016/j.aquaculture.2018.10.059.

[12] Y. Hao, K. Ding, Y. Xu, Y. Tang, D. Liu, G. Li, States, trends, and future of aquaponics research, Sustainability 12 (18) (2020) 7783, https://doi.org/10.3390/su12187783.

[13] B. Yep, Y. Zheng, Aquaponic trends and challenges—A review, J. Clean. Prod. 228 (2019) 1586–1599, https://doi.org/10.1016/j.jclepro.2019.04.290.

[14] Y.-J. Wang, T. Yang, H.-J. Kim, pH dynamics in aquaponic systems: implications for plant and fish crop productivity and yield, Sustainability 15 (9) (2023) 7137, https://doi.org/10.3390/su15097137.

[15] W.R. Beauchamp, J.M. Pickens, J.L. Sibley, J.A. Chappell, N.R. Martin, A.F. Newby, Salt level in a simulated aquaponic system and effects on Bibb lettuce, Int. J. Veg. Sci. 24 (2) (2018) 122–136, https://doi.org/10.1080/19315260.2017.1378787.

[16] J. Prüter, S.M. Strauch, L.C. Wenzel, W. Klysubun, H.W. Palm, P. Leinweber, Organic matter composition and phosphorus speciation of solid waste from an African catfish recirculating aquaculture system, Agriculture 10 (10) (2020) 466, https://doi.org/10.3390/agriculture10100466.

[17] N.A. Savidov, E. Hutchings, J.E. Rakocy, Fish and plant production in a recirculating aquaponic system: a new approach to sustainable agriculture in Canada, Acta Hortic. 742 (2007) 209–221, https://doi.org/10.17660/ActaHortic.2007.742.28.

[18] L. Harabiš, M. Šorf, M. Radojičić, J. Mareš, Optimization of aerobic digestion conditions of solid waste from recirculating aquaculture systems for liquid fertilizer production, Aquac. Eng. 111 (2025) 102574, https://doi.org/10.1016/j.aquaeng.2025.102574.

[19] B. Delaide, H. Monsees, A. Gross, S. Goddek, Aerobic and anaerobic treatments for aquaponic sludge reduction and mineralisation, in: S. Goddek, A. Joyce, B. Kotzen, G.M. Burnell (Eds.), Aquaponics Food Production Systems: Combined Aquaculture and Hydroponic Production Technologies for the Future, Springer, 2019, pp. 247–266, https://doi.org/10.1007/978-3-030-15943-6_10.

[20] S. Goddek, B.P. Delaide, A. Joyce, S. Wuertz, M.H. Jijakli, A. Gross, E.H. Eding, I. Bläser, M. Reuter, L.P. Keizer, Nutrient mineralization and organic matter reduction performance of RAS-based sludge in sequential UASB-EGSB reactors, Aquac. Eng. 83 (2018) 1–19, https://doi.org/10.1016/j.aquaeng.2018.07.003.

[21] R.M. Guyapale, C. Dionela, F. Huervana, Comparison of the nutrient composition of sludge under aerobic and anaerobic mineralization from African catfish, *Clarias gariepinus* (Actinopterygii: siluriformes: clariidae), reared in an intensive recirculating aquaculture system, Acta Ichthyol. Piscatoria 54 (2024) 275–282, https://doi.org/10.3897/aiep.54.131982.

[22] H. Monsees, J. Keitel, M. Paul, W. Kloas, S. Wuertz, Potential of aquacultural sludge treatment for aquaponics: evaluation of nutrient mobilization under aerobic and anaerobic conditions, Aquac. Environ. Interact. 9 (2017) 9–18, https://doi.org/10.3354/aei00205.

[23] J.E. Rakocy, D.S. Bailey, R.C. Shultz, J.J. Danaher, Preliminary evaluation of organic waste from two aquaculture systems as a source of inorganic nutrients for hydroponics, Acta Hortic. 742 (2007) 201–208, https://doi.org/10.17660/ActaHortic.2007.742.27.

[24] H. Zhang, Y. Gao, H. Shi, C.T. Lee, H. Hashim, Z. Zhang, W.-M. Wu, C. Li, Recovery of nutrients from fish sludge in an aquaponic system using biological aerated filters with ceramsite plus lignocellulosic material media, J. Clean. Prod. 258 (2020) 120886, https://doi.org/10.1016/j.jclepro.2020.120886.

[25] V.P. Lobanov, D. Combot, P. Pelissier, L. Labbé, A. Joyce, Improving plant health through nutrient remineralization in aquaponic systems, Front. Plant Sci. 12 (2021) 683690, https://doi.org/10.3389/fpls.2021.683690.

[26] M. Ezziddine, H. Liltved, R. Seljåsen, Hydroponic lettuce cultivation using organic nutrient solution from aerobic digested aquacultural sludge, Agronomy 11 (8) (2021) 1484, https://doi.org/10.3390/agronomy11081484.

[27] B. Delaide, S. Goddek, J. Gott, H. Soyeurt, M.H. Jijakli, Lettuce (*Lactuca sativa* L.) growth performance in complemented aquaponic solution outperforms hydroponics, Water 8 (10) (2016) 467, https://doi.org/10.3390/w8100467.

[28] S. Khalil, P. Panda, F. Ghadamgahi, A.-K. Rosberg, R.R. Vetukuri, Comparison of two commercial recirculated aquacultural systems and their microbial potential in plant disease suppression, BMC Microbiol. 21 (2021) 125, https://doi.org/10.1186/s12866-021-02273-4.

[29] E. Banchio, X. Xie, H. Zhang, P.W. Paré, Soil bacteria elevate essential oil accumulation and emissions in sweet basil, J. Agric. Food Chem. 57 (16) (2009) 6531–6536, https://doi.org/10.1021/jf8020305.

[30] J.S. Mangmang, R. Deaker, G. Rogers, Inoculation effect of *Azospirillum brasilense* on basil grown under aquaponics production system, Org. Agric 6 (1) (2016) 65–74, https://doi.org/10.1007/s13165-015-0115-5.

[31] O.J. Afolabi, O.O. Oladele, F.C. Olususi, Assessment of bacterial loads of *Clarias gariepinus* (Burchell, 1822) obtained from cultured and natural habitats, J. Basic Appl. Zool. 81 (1) (2020) 32, https://doi.org/10.1186/s41936-020-00168-w.

[32] D.S.Y.R. Putri, D.H. Achhlam, S. Andriyono, D. Hariani, H.D.K. Do, S.P. A. Wahyuningsih, Supplementation of *Bacillus subtilis* and *Lactobacillus casei* to increase growth performance and immune system of catfish (*Clarias gariepinus*) due to *Aeromonas hydrophila* infection, Vet. World 17 (3) (2024) 602–611, https://doi.org/10.14202/vetworld.2024.602-611.

[33] J. Clols-Fuentes, J.A. Nguinkal, P. Unger, B. Kreikemeyer, H.W. Palm, Bacterial community in African catfish (*Clarias gariepinus*) recirculating aquaculture systems under different stocking densities, Front. Mar. Sci. 10 (2023) 1073250, https://doi.org/10.3389/fmars.2023.1073250.

[34] Food and Agriculture Organization of the United Nations, The State of World Fisheries and Aquaculture 2024: Blue Transformation in Action, FAO, 2024, https://doi.org/10.4060/cd0683en.

[35] P. Suppakul, J. Miltz, K. Sonneveld, S.W. Bigger, Antimicrobial properties of basil and its possible application in food packaging, J. Agric. Food Chem. 51 (11) (2003) 3197–3207, https://doi.org/10.1021/jf021038t.

[36] A. de S. Botelho, O.O. Ferreira, M.S. de Oliveira, J.N. Cruz, S.H. Chaves, R. dos, A. F. do Prado, L.D. do Nascimento, G.A. da Silva, C.B. do Amarante, E.H. de Aguiar Andrade, Studies on the phytochemical profile of *Ocimum basilicum* var. *minimum* (L.) alef. essential oil, its larvicidal activity, and in silico interaction with acetylcholinesterase against *Aedes aegypti* (Diptera: culicidae), Int. J. Mol. Sci. 23 (19) (2022) 11172, https://doi.org/10.3390/ijms231911172.

[37] Á. Rodríguez-González, S. Álvarez-García, Ó. González-López, F. Da Silva, P. A. Casquero, Insecticidal properties of *Ocimum basilicum* and *Cymbopogon winterianus* against *Acanthoscelides obtectus*, insect pest of the common bean (*Phaseolus vulgaris*, L.), Insects 10 (5) (2019) 151, https://doi.org/10.3390/insects10050151.

[38] A. Rajali, N. Zain, N. Amran, N.E.M. Azmi, Antifungal efficacy of *Ocimum basilicum* essential oil in tissue conditioner against *Candida albicans*: an in vitro study, Contemp. Clin. Dent. 14 (2) (2023), https://doi.org/10.4103/ccd.ccd_654_21.

[39] E.B. Braun-Howland, T.E. Baxter, W.C. Lipps (Eds.), Standard Methods for the Examination of Water and Wastewater, 24th ed., APHA Press, 2023.

[40] Z. Zloch, J. Čelakovský, A. Aujezdská, Stanovení Obsahu Polyfenolů a Celkové Antioxidační Kapacity V Potravinách Rostlinného Původu, Ústav hygieny Lékařské fakulty UK, 2004.

[41] Zbíral, J. (Ed.). (2005). Analýza Rostlinného Materiálu: Jednotné Pracovní Postupy. ÚKZÚZ, Národní referenční laboratoř.

[42] G. Holm, Chlorophyll mutations in barley, Acta Agric. Scand. 4 (1954) 457–471, https://doi.org/10.1080/00015125409439955.

[43] L.F. Posada, J.C. Alvarez, C.-H. Hu, L.E. de-Bashan, Y. Bashan, Construction of probe of the plant growth-promoting bacteria *Bacillus subtilis* useful for fluorescence in situ hybridization, J. Microbiol. Methods 127 (2016) 1–8, https://doi.org/10.1016/j.mimet.2016.05.029.

[44] M. Stoffels, T. Castellanos, A. Hartmann, Design and application of new 16S rRNA-targeted oligonucleotide probes for the azospirillum-skermanella-rhodocista-cluster, Syst. Appl. Microbiol. 24 (2001) 83–97, https://doi.org/10.1078/0723-2020-00011.

[45] C. Alarcón, G. Cuenca, Arbuscular mycorrhizas in coastal sand dunes of the Paraguaná Peninsula, Venezuela, Mycorrhiza 16 (1) (2005) 1–9, https://doi.org/10.1007/s00572-005-0005-x.
[46] H. Koch, M.A. van Kessel, S. Lücker, Complete nitrification: insights into the ecophysiology of comammox *Nitrospira*, Appl. Microbiol. Biotechnol. 103 (2019) 177–189, https://doi.org/10.1007/s00253-018-9486-3.
[47] X. Bai, F. Hazi, I. Takacs, T. Wadhawan, W.J. Parker, A comprehensive floc model for simulating simultaneous nitrification, denitrification, and phosphorus removal, Sci. Total Environ. 927 (2024) 172023, https://doi.org/10.1016/j.scitotenv.2024.172023.
[48] S. Ahn, M. Cho, M.J. Sadowsky, J. Jang, Dissimilatory nitrate reductions in soil *Neobacillus* and *Bacillus* strains under aerobic condition, J. Microbiol. 63 (2) (2025) e2411019, https://doi.org/10.71150/jm.2411019.
[49] B.R. Giri, S. Chattaraj, S. Rath, M.M. Pattnaik, D. Mitra, H. Thatoi, Unveiling the molecular mechanism of *Azospirillum* in plant growth promotion, Bacteria 4 (3) (2025) 36, https://doi.org/10.3390/bacteria4030036.
[50] E. Solis-Toapanta, P. Fischer, C. Gómez, Growth rate and nutrient uptake of basil in small-scale hydroponics, Hortscience 55 (4) (2020) 507–514, https://doi.org/10.21273/HORTSCI14727-19.
[51] B. Prinsi, N. Negrini, S. Morgutti, L. Espen, Nitrogen starvation and nitrate or ammonium availability differently affect phenolic composition in green and purple basil, Agronomy 10 (4) (2020) 498, https://doi.org/10.3390/agronomy10040498.
[52] D.P. Gillespie, C. Kubota, S.A. Miller, Effects of low pH of hydroponic nutrient solution on plant growth, nutrient uptake, and root rot disease incidence of basil (*Ocimum basilicum* L.), Hortscience 55 (8) (2020) 1251–1258, https://doi.org/10.21273/HORTSCI14986-20.
[53] B. Delaide, E. Panana, S. Teerlinck, P. Bleyaert, Suitability of supernatant of aerobic and anaerobic pikeperch (*Sander lucioperca* L.) sludge treatments as a water source for hydroponic production of lettuce (*Lactuca sativa* L. var. *capitata*), Aquac. Int. 29 (4) (2021) 1721–1735, https://doi.org/10.1007/s10499-021-00709-2.
[54] S. Goddek, Z. Schmautz, B. Scott, B. Delaide, K.J. Keesman, S. Wuertz, R. Junge, The effect of anaerobic and aerobic fish sludge supernatant on hydroponic lettuce, Agronomy 6 (2) (2016) 37, https://doi.org/10.3390/agronomy6020037.
[55] A.A. Tellbüscher, R. Gebauer, M. Šeda, T. Goldhammer, O. Nikl, K. Roy, W. Kloas, J. Mráz, Fish manure as a potential feedstock for organic hydroponic nutrient solutions: the effect of fish feed composition and fertilizer amendment on the growth performance of lettuce, Sci. Hortic. 350 (2025) 114309, https://doi.org/10.1016/j.scienta.2025.114309.
[56] D.T. Britto, H.J. Kronzucker, Nitrogen acquisition, PEP carboxylase, and cellular pH homeostasis: new views on old paradigms, Plant Cell Environ. 28 (11) (2005) 1396–1409, https://doi.org/10.1111/j.1365-3040.2005.01372.x.
[57] Y. Zou, Z. Hu, J. Zhang, H. Xie, C. Guimbaud, Y. Fang, Effects of pH on nitrogen transformations in media-based aquaponics, Bioresour. Technol. 210 (2016) 81–87, https://doi.org/10.1016/j.biortech.2015.12.079.
[58] N. Kasozi, B. Abraham, H. Kaiser, B. Wilhelmi, The complex microbiome in aquaponics: significance of the bacterial ecosystem, Ann. Microbiol. 71 (2021) 1, https://doi.org/10.1186/s13213-020-01613-5.
[59] S.E. Wortman, Crop physiological response to nutrient solution electrical conductivity and pH in an ebb-and-flow hydroponic system, Sci. Hortic. 194 (2015) 34–42, https://doi.org/10.1016/j.scienta.2015.07.045.
[60] H. Hosseini, V. Mozafari, H.R. Roosta, H. Shirani, P.C. Vlasakker, M. Farhangi, Nutrient use in vertical farming: optimal electrical conductivity of nutrient solution for growth of lettuce and basil in hydroponic cultivation, Horticulturae 7 (9) (2021) 283, https://doi.org/10.3390/horticulturae7090283.
[61] S. Goddek, T. Vermeulen, Comparison of *Lactuca sativa* growth performance in conventional and RAS-based hydroponic systems, Aquac. Int. 26 (2018) 1377–1386, https://doi.org/10.1007/s10499-018-0293-8.
[62] P. Marschner, Marschner's Mineral Nutrition of Higher Plants, third ed., Academic Press, 2012 https://doi.org/10.1016/C2009-0-63043-9.
[63] R.P. Germano, S. Melito, S. Cacini, G. Carmassi, F. Leoni, R. Maggini, F. F. Montesano, A. Pardossi, D. Massa, Sweet basil can be grown hydroponically at low phosphorus and high sodium chloride concentration: effect on plant and nutrient solution management, Sci. Hortic. 304 (2022) 111324, https://doi.org/10.1016/j.scienta.2022.111324.
[64] C.J. Currey, *Nutrient antagonism—The potassium-calcium-magnesium relationship* (e-GRO alert no. E614). e-GRO. https://www.e-gro.org/pdf/E614.pdf, 2021.
[65] D.P. Gillespie, G. Papio, C. Kubota, High nutrient concentrations of hydroponic solution can improve growth and nutrient uptake of spinach (*Spinacia oleracea* L.) grown in acidic nutrient solution, Hortscience 56 (6) (2021) 687–694, https://doi.org/10.21273/HORTSCI15777-21.
[66] J.K. Mercês, M.J. Medelo, A.B. Filho, Biofortification and quality of collard greens as a function of iron concentration in nutrient solution, Agronomy 12 (10) (2022) 2493, https://doi.org/10.3390/agronomy12102493.
[67] L. Signorini, G.C. Modarelli, P.D. Pierro, A.L. Langellotti, C. Cirillo, S.D. Pascale, P. Masi, Effects of seedling substrate and hydroponic versus aquaponic nutrient solution on growth, nutrient uptake, and eco-physiological response of lemon basil (*Ocimum* × *citriodorum*), Plants 14 (13) (2025) 1929, https://doi.org/10.3390/plants14131929.
[68] H. Attia, F. Rebah, C. Ouhibi, M.A. Saleh, A.T. Althobaiti, K.H. Alamer, M.B. Nasri, M. Lachaâl, Effect of potassium deficiency on physiological responses and anatomical structure of basil, *Ocimum basilicum* L, Biology 11 (11) (2022) 1557, https://doi.org/10.3390/biology11111557.
[69] A. Mendrinna, S. Persson, Root hair growth: it's a one way street, F1000Prime Rep. 7 (2015) 23, https://doi.org/10.12703/P7-23.
[70] M.K. Ashley, M. Grant, A. Grabov, Plant responses to potassium deficiencies: a role for potassium transport proteins, J. Exp. Bot. 57 (2) (2006) 425–436, https://doi.org/10.1093/jxb/erj034.
[71] X.Z. Fang, X.X. Liu, Y.X. Zhu, J.Y. Ye, C.W. Jin, The K^+ and NO_3^- interaction mediated by NITRATE TRANSPORTER1.1 ensures better plant growth under K^+-limiting conditions, Plant Physiol. 184 (4) (2020) 1900–1916, https://doi.org/10.1104/pp.20.01229.
[72] J.E. Rakocy, M.P. Masser, T.M. Losordo, *Recirculating Aquaculture Tank Production Systems: Aquaponics—Integrating Fish and Plant Culture* (SRAC Publication No. 454), Southern Regional Aquaculture Center, 2006.
[73] A. Mourantian, M. Aslanidou, E. Mente, N. Katsoulas, E. Levizou, Basil functional and growth responses when cultivated via different aquaponic and hydroponics systems, PeerJ 11 (2023) e15664, https://doi.org/10.7717/peerj.15664.
[74] K. Patloková, R. Pokluda, Optimization of plant nutrition in aquaponics: the impact of *Trichoderma harzianum* and *Bacillus mojavensis* on lettuce and basil yield and mineral status, Plants 13 (2) (2024) 291, https://doi.org/10.3390/plants13020291.
[75] National Center for Biotechnology Information, BLAST: basic local alignment search tool, from, https://blast.ncbi.nlm.nih.gov/Blast.cgi, 2025. (Accessed 21 December 2025).
[76] M. Ruzzi, R. Aroca, Plant growth-promoting rhizobacteria act as biostimulants in horticulture, Sci. Hortic. 196 (2015) 124–134, https://doi.org/10.1016/j.scienta.2015.08.042.
[77] D. Rodgers, E. Won, M.B. Timmons, N. Mattson, Complementary nutrients in decoupled aquaponics enhance basil performance, Horticulturae 8 (2) (2022) 111, https://doi.org/10.3390/horticulturae8020111.
[78] E. Skvortsova, O. Filinskaya, I. Postrash, A. Bushkareva, A. Mostofina, Biodiversity of gut microorganisms in aquacultured African catfish, E3S Web Conf. 463 (2023) 1039, https://doi.org/10.1051/e3sconf/202346301039.