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Anthroponics: Application and effects on growth of parsley, rhipsalis, coriander, and basil fed with urine fertiliser



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ABSTRACT

Urine has emerged as a promising nutrient-rich waste stream suitable for plant fertigation. Biological nitrification in a membrane bioreactor offers an effective method for producing liquid fertiliser from urine, extracting essential nutrients into a final product. Despite the increasing interest in hydroponics and urban horticulture, research exploring the efficacy of urine-derived fertilisers remains scarce. This study employed three growing systems, including nutrient film technique open channels in outdoor setup, pot-drench under glasshouse conditions, and deep-water culture (DWC) under laboratory conditions to investigate the efficiency of urine-derived fertiliser compared to commercial fertilisers. The results demonstrated comparable growth responses in parsley, rhipsalis, coriander, and basil, as demonstrated by the analysis of several parameters including biomass, stem numbers and length, and nutritional composition in shoots and roots. In conclusion, urine-derived fertilisers showed promising potential as a sustainable alternative to synthetic fertilisers, laying the foundation for the development of a circular economy of nutrients in agriculture.

1. Introduction

The sustainable cultivation of crops has been an ever-pressing concern in the face of the increasing global population and the concomitant demand for food. Further, the crop production is projected to have a 49% to 117% increase by 2100 according to 2005 levels, since the future population is expected to increase from 7.3 billion to 12.6 billion by the end of this century [1–3]. Conventional agricultural practices have relied on synthetic fertilisers, which not only deplete finite resources but also consume 1–2% of total world energy demand by Haber-Bosch process and pose environmental challenges due to nutrient runoff [4]. Moreover, Russia's termination of fertiliser export during the 2022 Russia-Ukraine war caused fertiliser shortages and soaring costs worldwide [5]. In this context, pathways to a circular economy of nutrients have gained significant prominence. The circular economy promotes the efficient and sustainable use of resources, where waste is minimised, and nutrients are continuously recycled and reused [6–8].

The introduction of circular economy strategies in agriculture entails the recycling of nutrients. Nutrient recovery from waste streams offers an opportunity to minimise the environmental footprint of agriculture while ensuring the efficient use of essential elements like nitrogen and phosphorous [9–12]. This approach involves recycling the extracted nutrients as fertilisers, consequently diminishing reliance on synthetic fertilisers and alleviating their adverse environmental impacts. Among the plethora of nutrient-rich waste streams, human urine has emerged as an intriguing candidate. Urine, often considered a waste product, contains valuable nutrients at high concentrations, including nitrogen (3-4 g/L) and phosphorus (0.3–0.6 g/L), which are essential for plant growth [13,14]. The practice of source-separating urine and reclaiming nutrients from it offers a distinctive opportunity to develop sustainable fertilisers underpinned by a circular economy vision [15]. An increasing number of studies worldwide have highlighted the value of urine as a fertiliser for crops such as tomatoes and wheat, demonstrating its high feasibility and effectiveness in agricultural application [16,17].

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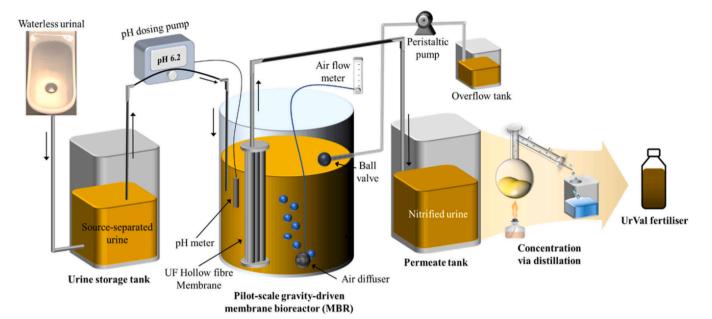


Fig. 1. Schematic diagram of UrVal fertiliser production process.

Table 1
Composition of treatment solutions used in case I, II, and III.

Concentration of treatment solution	Case I			Case II		Case III		
	T1_1 Commercial fertiliser	T2_1 UrVal	T3_1 Fortified UrVal	T1_2 Ca-Mg Grower	T2_2 UrVal	T1_3 Commercial fertiliser	T2_3 UrVal	T3_3 UrVal with high nitrite
Total Nitrogen (mg/L)	172.4	175	175	100.5	100.75	348	350	356
-Ammonium (mg/L)	n.a	n.a	n.a	n.a	n.a	140	180	140
-Nitrate (mg/L)	n.a	n.a	n.a	n.a	n.a	208	170	96
-Nitrite (mg/L)	n.a	n.a	n.a	n.a	n.a	0	0	120
Phosphorus (mg/L)	32	7.35	7.35	14.74	6.83	10.2	6.2	5.6
Potassium (mg/L)	240.4	52.5	52.5	83.08	48.75	355.4	109.7	79.5
Calcium (mg/L)	144	1.4	71.4	33.5	1.3	36	2.81	2.52
Magnesium (mg/L)	40	0.14	21.14	12.06	0.13	84	9.4	3.7
Sulfur (mg/L)	52.8	5.6	6.76	0	5.2	110	90	85
Iron (mg/L)	1.76	0.0018	2.70	0.80	0.0016	2.54	0.04	0.02
Copper (mg/L)	0.12	0.0011	0.0056	0.1005	0.0010	0.1	0.002	0.002
Zinc (mg/L)	0.16	0.0525	0.0683	0.1005	0.0488	0.1	0.08	0.08
Boron (mg/L)	0.24	0.056	0.231	0.134	0.052	0.2	0.08	0.08
Manganese (mg/L)	0.72	0.34	0.50	0.40	0.31	0.58	0.18	0
Molybdenum (mg/L)	0.04	0.007	0.011	0.067	0.007	0.05	0.01	0.01
Conductivity (µS/cm)	1508	488	582	n.a	n.a	1392	1396	1401

^{*}n.a: not analysed

Nonetheless, the direct application of urine as a plant fertiliser poses inherent challenges, including elevated levels of ammonia and pH, resulting in ammonia volatilization, as well as high salinity (1.1–1.7 g/L), high organic content up to 5 g/L of chemical oxygen demand (COD), and potential contaminants such as pathogens and viruses. The implementation of urine nitrification within a membrane bioreactor (MBR) has emerged as a highly promising strategy for the production of liquid fertiliser that retains all essential nutrients in a singular end product [13]. The biological oxidation of half the ammonia to nitrate not only facilitates pH stabilisation without the need for chemical additives but also contributes to the removal of organic matter [18,19]. Furthermore, the incorporation of an ultrafiltration (UF) membrane enables the exclusion of viruses and pathogens, ensuring the production of a safe and efficacious liquid fertiliser [20,21].

Hydroponic gardening, which is a soilless culture system, has emerged as an alternative to traditional field farming, characterised by its high efficiency in water and nutrient use for plant cultivation. This technique minimises the risk of plant pests and diseases due to the independence

from soil conditions and enables higher crop yields per area unit compared to conventional horizontal farming [22]. For instance, hydroponic lettuce production resulted in 11 times greater yield than conventional agricultural method [23]. Therefore, liquid fertilisers are critical to ensure the desired functioning of hydroponic systems, providing nutrients, water, and oxygen to plants [24]. While hydroponics have been used in traditional large-scale production of vegetables during the last three to four decades, this technique is now moving closer to the consumers and is available for applications in all types of households, including city apartments [25]. The innovative concept of anthroponics represents a pivotal convergence of sustainable urban agriculture and responsible waste management. Anthroponics, a subset of hydroponics, utilises human urine as a primary nutrient source for plant cultivation [26]. This approach reduces environmental pollution associated with synthetic fertiliser production and decreases dependency on these fertilisers. The application of anthroponics can bring about the possibilities of local closed-loop nutrient cycles by recovering nutrients from human waste and implementing hydroponic plant fertigation. Recent studies have increasingly focused on proof-of-

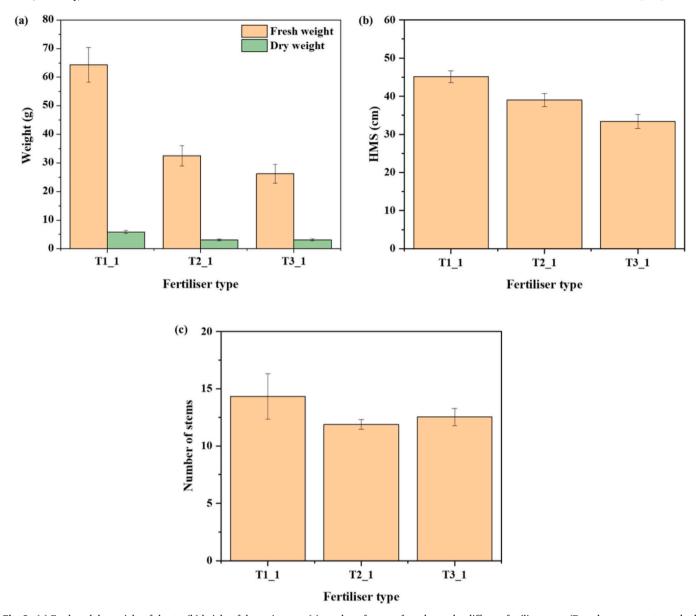


Fig. 2. (a) Fresh and dry weight of shoots; (b) height of the main stem; (c) number of stems of parsley under different fertiliser types (Error bars represent standard error of replications).

concept systems for anthroponics, demonstrating its potential for local urine treatment and reuse in domestic food production, particularly in urban settings [27–29]. Volpin et al. (2020) have highlighted the promising performance of urine-derived fertiliser in growing lettuce and pak choi, showing comparable results to commercial fertilisers in vertical hydroponic gardens [30]. However, given the distinctive nutrient requirements of different plant species within hydroponic environments, growth responses may diverge. Therefore, conducting comprehensive studies to assess the efficacy of urine fertilisers and its formulation is imperative for fostering widespread adoption in anthroponic systems.

This study focused on quantifying the effectiveness of a urine-derived fertiliser obtained from the biological nitrification in a UF-MBR system. By selecting different growing set-ups and type of plants, three case studies were carried out.

 Case I aimed to investigate the effects of urine fertiliser on the growth of Parsley (*Petroselinum crispum*) in an outdoor open gutters' nutrient film technique (NFT) module' [31], in comparison to a commercial fertiliser and a fortified urine solution with added micronutrients.

- Case II focused on the effects of urine fertiliser on the growth of Rhipsalis teres (Vell.) Steud under controlled glasshouse conditions compared to a commercial fertiliser.
- Case III investigated the effect of high-nitrite concentration urine fertiliser, a commercial fertiliser and zero-nitrite urine fertiliser on the growth of coriander (*Coriandrum sativum*) and basil (*Ocimum basilicum*) in a deep-water culture unit (Hydrogarden). Since there remains a lack of research on the impact of high nitrite concentration on plants in hydroponic applications, this trial aimed to address this knowledge gap.

This study unravelled the role of anthroponics as a novel approach to urban agriculture and detail the integration of human waste into the food production cycle.

2. Materials and methods

2.1. Production of UrVal fertiliser

A 60 L pilot-scale gravity-driven membrane bioreactor (MBR) was employed for the nitrification of urine. The seed sludge was obtained

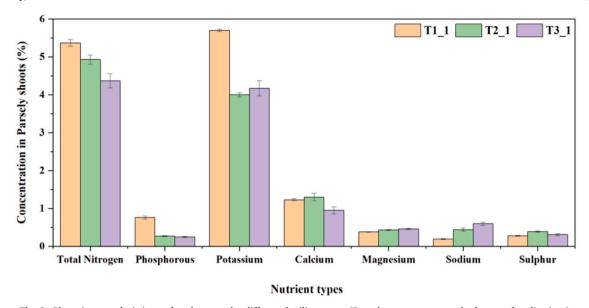


Fig. 3. Plant tissue analysis in parsley shoots under different fertiliser types (Error bars represent standard error of replications).

from a decentralized wastewater treatment plant situated in Central Park, Sydney, Australia, and was subsequently acclimated with diluted urine, gradually transitioning to normal urine concentration. Source-separated urine was conveniently collected from the urinal room in Building 11 at the University of Technology Sydney and stored in a 100 L water tank for complete urea hydrolysis. Throughout the MBR operation, a pH meter (HI6100405, Hanna Instruments, Australia) was interfaced with a pH-controlling dosing pump (BL7916-1, Hanna Instruments, Australia) to ensure pH stability at 6.2 which is crucial for optimising the nitrification process in the MBR [30]. This was achieved by automatically introducing high pH urine (pH 9.2) whenever a decrease in pH occurred in the bioreactor due to alkalinity consumption. Additionally, a commercially available polyvinylidene fluoride (PVDF) hollow fibre ultrafiltration membrane module (Beijing OriginWater Technology, China) with a pore size of 0.02 μm , and active surface area of 0.06 m^2 was utilized in this study. Furthermore, a floating ball valve was installed on the sidewall of the MBR to serve as a water level controller, maintaining the constant hydrostatic pressure necessary for ultrafiltration, as well as avoid any overflow. Nitrified urine was collected from the permeate tank during stable operation and subsequently concentrated 20-fold using a commercial distiller to produce urine fertiliser, named UrVal fertiliser [15]. The details of this process, including the nitrification performance, can be found previous studies [32,33]. Fig. 1 illustrates the schematic diagram of the production process of UrVal fertiliser, from the biological nitrification in the MBR to concentration of nitrified urine via distillation.

2.2. Experimental set-up

Parsley, rhipsalis, coriander, and basil were selected for the study based on several criteria that make these plants suitable for evaluating the effectiveness of urine fertiliser. Firstly, these plants have rapid growth cycles, allowing for quicker observation of growth responses and nutrient uptake. Additionally, these species are well-researched regarding their nutrient requirements, providing a solid foundation for accurate interpretation of results. Moreover, these species have low to medium nutrient needs (100–200 ppm), which can be easily met by diluting urine fertilisers. Finally, these plants have significant potential for urban agriculture and horticulture, making them relevant for studies aimed at sustainable and practical agricultural solutions. The planting methods in the separate case studies were selected based on their suitability to replicate commercial horticultural practices. Across all case studies, urine fertiliser was benchmarked against various advanced commercial fertilisers with balanced nutrient compositions.

2.2.1. Case I - parsley growth in nutrient film technique (NFT) open channels

The nutrient film technique (NFT) system was selected, among others, due to its flexibility, versatility, and wide application in horticulture. The experiment was carried out for two months at the Royal Botanical Garden (RBG) Sydney Nursery in an outdoor open gutter NFT module (Fig. S1a). Parsley (*Petroselinum crispum*, Johnsons seeds, Australia) was germinated in plugs filled with a seed raising media, grown in a controlled temperature glasshouse (18–27°C) for about 3 weeks and fed with the respective nutrient solutions. Climatic data were recorded, summarised by a minimum temperature of 8 \pm 2°C, a maximum of 24 \pm 4°C and a 9 AM humidity of 67 \pm 10%.

The performance of a commercial hydroponic solution "T1 1" (Optimum Grow twin pack hydroponic nutrient, Growth Technology, Australia) was benchmarked with UrVal fertiliser as nutrient solution "T2 1" and a fortified urine solution (with the addition of 200 g Ca-EDTA/L, 100 g Mg-EDTA/L, 4.82 g Fe-EDTA/L and other micronutrients) as nutrient solution "T3_1" (see compositions in Table 1). The growing medium used was a 135 mm coco disc pellet (3.3 L, 10–30 mm, Garden City Plastics, Australia) which was placed in 200 mm plastic pots that were soaked in water until the medium had fully expanded. Three parsley seedlings were transplanted into a single 200 mm pot filled with coco pellets before being placed in the gutter. Each nutrient solution treatment consisted of a single row of ten 200 mm pots, totalling 30 plants each. Nutrient solutions were pumped (AquaPro Submersible pump, AP1050) from a 30 L container through a 13 mm low density polyethylene pipe which recirculated 300 L/h of nutrient solution to each treatment. Nutrient solutions drained by gravity from the top end of the gutter to the lower end (each row was $240\,\mathrm{cm}\,\mathrm{L}~\mathrm{x}$ $20\,\text{cm}\,\text{W}\,\text{x}\,2.5\,\text{cm}\,\text{D})$ through a $40\,\text{mm}$ drainage pipe back to the nutrient solution containers placed below the bottom end of each treatment. The recirculation allowed for sufficient dissolved oxygen [34] and water to be transferred to the growing medium by capillary rise. Each nutrient solution was diluted with deionised (DI) water to achieve a nitrogen concentration of about 170 mg N/L. Finally, the diluted solutions were replenished on a weekly basis to maintain the water level and electrical conductivity (EC), while the pH was adjusted using KOH or H_2PO_4 to keep it at 6.1 \pm 0.2 (TPS -Aqua CPA, V6819). The image of NFT open channels experimental set up is shown in Fig. S1a.

2.2.2. Case II - rhipsalis growth under controlled glasshouse conditions Rhipsalis (Rhipsalis teres (Vell.) Steud, RBG Sydney, Australia) was grown for three months at the RBG Sydney Nursery in a glasshouse with

controlled temperature (16–29 °C). The experiment was arranged in a complete randomized design (CRD) with 10 replications, as shown in Fig. S1b. In this study, a Ca-Mg grower (Peters Excel CalMag grower, ICL Specialty fertilisers) solution labelled "T1_2" was compared to the performance of UrVal fertiliser as nutrient solution "T2_2". T1_2 was selected because it is a sophisticated fertiliser known for its maximised nutrient absorbability and usability, achieved through an advanced chelating formula that includes chelated trace elements and calcium and magnesium. The plants were fertigated (pot drench) on a weekly basis with a nitrogen concentration of 100 mg N/L, and the accumulation of sodium was not observed in the potting media at the selected feeding rate and over the period of experiment. The image of rhipsalis growth experimental set up in glasshouse is shown in Fig. S1b.

2.2.3. Case III – coriander and Basil growth in domestic indoor hydroponic systems

Coriander (*Coriandrum sativum*, Mr Fothergill's seed, Australia) and basil (*Ocimum basilicum*, Mr Fothergill's seed, Australia) were cultivated for 3 months in deep water culture indoor hydroponic system, 'HydroGarden', at the environmental laboratory at the University of Technology Sydney. A total of 6 hydrogarden units were installed, 3

units per each plant type and each unit fed with a different fertiliser solution. Vermiculite served as a growing medium, complemented by built-in LED lighting for efficient illumination, programmed to operate on a daylight cycle for automatic on and off functionality. A commercial fertiliser labelled "T1_3" (HydroBoost liquid fertiliser, Mr. Fothergill's, Australia), was compared to evaluate the efficacy of UrVal fertiliser as a nutrient solution, labelled "T2_3", and a urine-based fertiliser with elevated nitrite concentration, denoted as nutrient solution "T3_3". T3_3 was produced through a deliberate destabilization of nitrification within the MBR via ammonia shock loading, resulting in significant nitrite accumulation by inhibiting nitrite-oxidizing bacteria [13]. The comparison of T2_3 and T3_3 to assess the impact of nitrite presence in the hydroponic system on plant growth was crucial, as nitrite, an intermediate product, is a major limiting factor that significantly restricts the reduction of hydraulic retention time in the MBR process. In contrast to the UrVal fertiliser, the distillation process for concentration was skipped for T3 3. Instead, it was directly applied after dilution immediately upon collection from the MBR process to prevent any potential loss of nitrite, given nitrite's relative instability [35]. Each nutrient solution was diluted with deionised (DI) water to attain a nitrogen concentration of approximately 350 mg TN/L, aligning with the

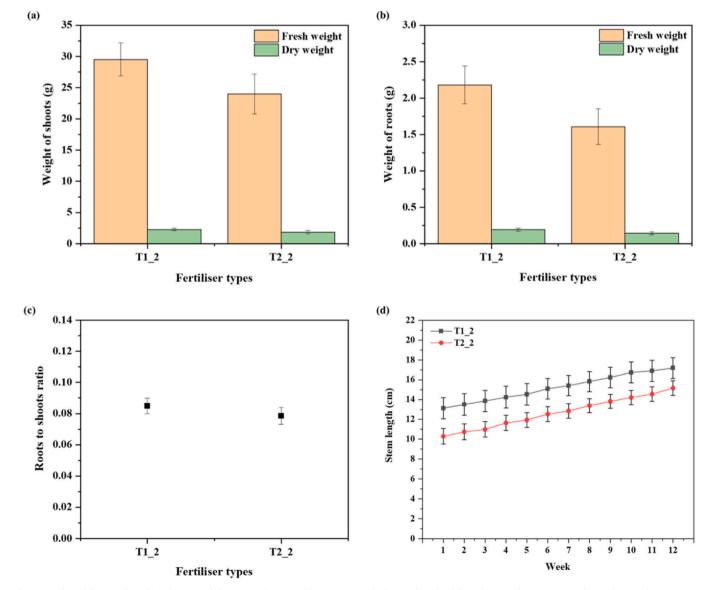


Fig. 4. Fresh and dry weights of (a) shoots; and (b) roots; (c) roots to shoots ratio; and (d) stem length of rhipsalis over the experimental period (Error bars represent standard error of replications).

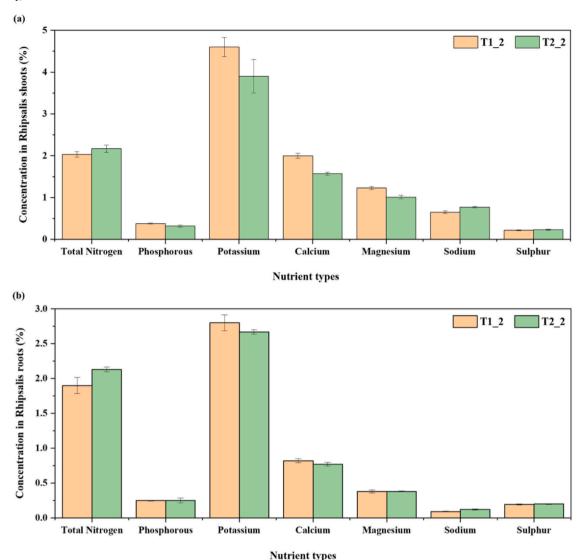


Fig. 5. Plant tissue analysis in rhipsalis (a) shoots; and (b) roots under different fertiliser types (Error bars represent standard error of replications).

formulation of the commercial fertiliser (T1_3). Germination of coriander and basil occurred after a two-week period, following which the respective nutrient solutions were replenished on a weekly basis. The image of experimental set up of hydrogardens is shown in Fig. S1c. The composition of all treatment solutions used in the three case studies is described in Table 1. Although T2_1, T2_2, and T2_3 are all UrVal fertilisers, the nutrient composition was not proportionally consistent across these variants due to the use of different batches from the MBR process in each case study.

2.3. Analysis methods

The treatment solution was collected from each container for each plant and diluted 10-fold for analysis. The ionic composition of each treatment solution was analysed using a standard test kit (Merck Millipore, Burlington, USA) and a photometer (Spectroquant NOVA 60, Merck, Germany) for anions, while cations were measured via Inductively coupled plasma mass spectrometry (ICP-MS).

At the end of each case study, the shoots and roots were separated and dried for subsequent dry weight measurement (refer to detailed procedure in Sections 2.3.1–2.3.3). Dried shoots and roots tissue samples were then sent to the New South Wales - Department of Primary Industries laboratory (Wollongbar laboratory, DPI, NSW, Australia) for plant tissue analysis to measure nutrient composition of shoots and

roots. Total Kjeldahl nitrogen, and 20 other elements were analysed using ICP, with nitrate nitrogen additionally analysed for case III. Full analysis results including micronutrients can be found in Fig. S2.

For data analysis, the paired t-test analysis was carried out in Microsoft Excel to determine the statistical significance of the data obtained. The significance of the differences between the treatments was considered at the 5% significance level ($p \le 0.05$).

2.3.1. Case I

The five containers from the middle of each gutter (treatment) were selected for data collection. The weight of samples was recorded using an EJ Series scale (AND, EJ-610, A&D Co LTD, Korea). The parsley shoots were separated to assess fresh biomass and dried in an oven at 60 $^{\circ}$ C for 72 h to assess the dry weight [34,36]. However, due to the difficulty in separating parsley roots from the medium, data collection for parsley roots was not conducted for case I. Height of Main Stem (HMS) was measured by a 50 cm ruler from the base to the tip of the tallest shoot. Stem number (SN) was measured by counting the number of stems branching from 5 cm above media level.

2.3.2. Case II

At the end of each experiment, the shoots and roots of each plant were separated to assess fresh biomass. The roots were separated from the growing media by gently washing them in a stream of water. Shoots

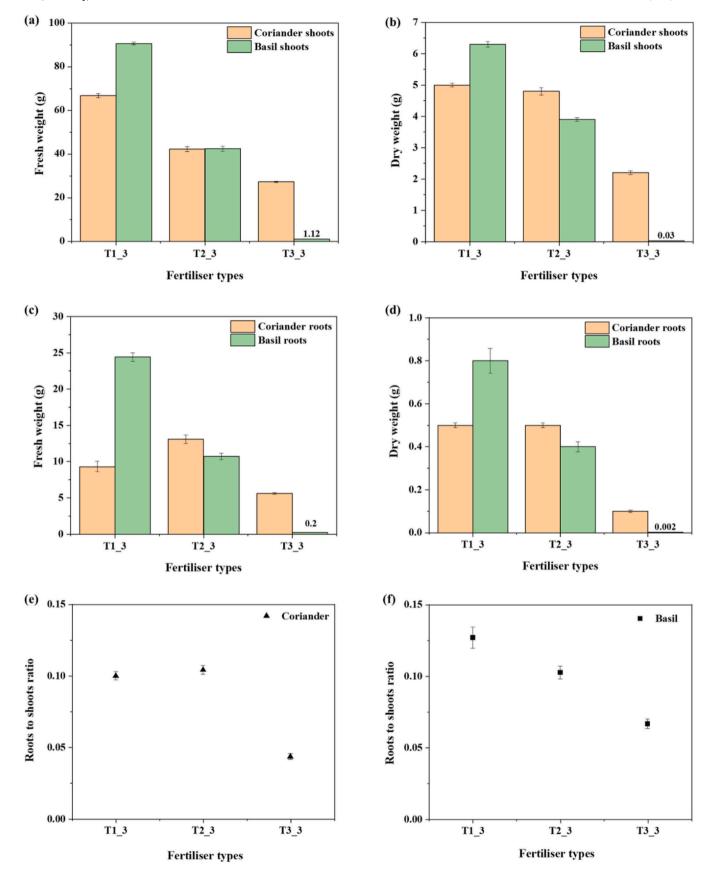


Fig. 6. (a) Fresh weight of shoots; (b) dry weight of shoots; (c) fresh weight of roots; (d) dry weight of roots; (e) roots to shoots ratio of coriander; and (f) roots to shoots ratio of basil (Error bars represent standard error of replications).

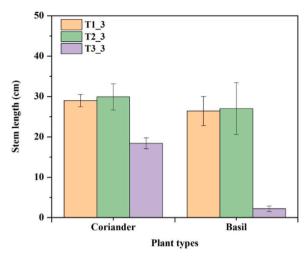


Fig. 7. Stem length of coriander and basil (Error bars represent standard error of replications).

and roots samples were placed in paper bags and dried in an oven at 60 \mathbb{C} for $72\,h$ [34,36]. Subsequently, they were weighed again to determine their dry weight. The roots-to-shoots ratio was calculated as the ratio between the dry weight of roots and the dry weight of shoots. The stem length data of Rhipsalis plants were collected at weekly intervals and over a 12 weeks period using a 30 cm ruler by measuring the longest stem from the base of the plant to the tip of the stem.

2.3.3. Case III

At the end of the experiment, data on the biomass of coriander and basil shoots and roots were collected, both in their fresh and oven-dried states, following the same methodology as in case II. Roots to shoots ratio was also calculated as the ratio between roots dry weight and shoots dry weight. Additionally, stem length of each plant was measured using a 50 cm ruler from the base to the tip of each shoot.

3. Results and discussion

3.1. Case I - parsley growth in nutrient film technique (NFT) open channels

Fig. 2 shows the response of parsley grown in NFT open channel system in terms of fresh and dry weights of parsley shoots, the height of the main stem (HMS), and the number of stems (SN) under three different fertilisers treatments. T1_1 indicates the commercial fertiliser solution, while T2_1 and T3_1 represents the UrVal fertiliser and fortified UrVal fertiliser, respectively. Overall, the growth vigour and performance across all treatments were notably robust, with T1_1 demonstrating the highest yield, followed by T2_1 and then T3_1. Notably, variability in parsley growth in this trial primarily manifested in higher biomass and stem height rather than branching. The mean HMS showed a significant difference among the three treatments, with the highest recorded for T1_1 (45 cm), followed by 39 cm for T2_1 and 33 cm for T3_1 (Fig. 2b) ($p \le 0.05$). Furthermore, significant variability in results was observed in terms of biomass fresh weight and dry weight, with T1_1 plants (64.3 g, 5.9 g) exhibiting approximately double the size of those from T2_1 (32.5 g, 3.1 g) and T3_1 (26.2 g, 3.1 g) ($p \le 0.05$). However, the difference in both fresh and dry weights between T2_1 and T3_1 was not obvious (p > 0.05). While the branching patterns, in terms of SN, appeared comparable, with T1_1 showing slightly higher average SN than the other two treatments (Fig. 2c), it is worth noting that their variances are insignificant among treatments (p > 0.05). In summary, while the yield achieved with pure urine fertiliser was lower compared to commercial fertiliser, the number of stems per plant remained consistent across treatments. The profitability of herb production hinges greatly on

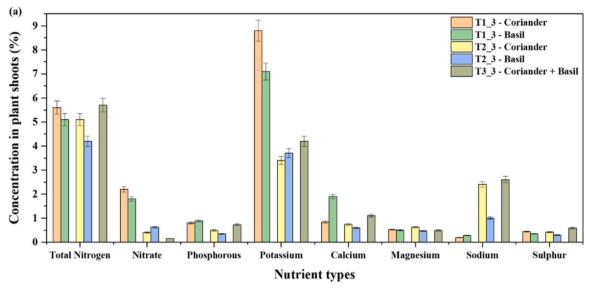
market dynamics, with bunch sizes primarily influenced by branch number per plant rather than biomass [37,38], suggesting promising prospects for nutrient solutions derived from recycled human urine. Further extensive trials with urine fertiliser are warranted to investigate the appropriate horticultural technology for herb production and to understand if higher yields could be achieved by simple process optimisation. The electrical conductivity (EC) values of urine nutrient solution (T2_1), as shown in Table 1, were three times lower than those of the commercial hydroponic fertiliser (i.e., 488 vs $1508\,\mu\text{S}/$ cm) due to the high dilution rate required to lower the nitrogen concentration. EC is a critical parameter as it can potentially result in soil salination and run-off to groundwater leading to the eutrophication. According to the Department of Environment and Conservation in NSW Australia, the EC in soil should be less than 2 mS/cm to avoid restricting plant growth and less than 4 mS/cm to prevent risk to groundwater [39]. In this context, urine fertiliser, characterised by a lower salt index, can be advantageous in mitigating environmental risks [30].

Fig. 3 presents the plant tissue analysis of parsley shoots in terms of major nutrients (N, P, K) and micronutrients (Ca, Mg, Na, S). The commercial T1_1 solution exhibited the highest content of major nutrients in parsley leaves and stems among the treatments, with total nitrogen, phosphorus, and potassium concentrations of 5.4%, 0.8%, and 5.7%, respectively ($p \le 0.05$). Despite the same total nitrogen concentration in the treatment solutions T2_1 and T3_1, their contents in both solutions were marginally lower than that of the commercial treatment ($p \le 0.05$), with values of 4.9% and 4.4% respectively. While phosphorus (P) and potassium (K) concentrations were approximately 4-5 times lower in UrVal fertilisers (T2_1 and T3_1) compared to T1_1, their contents in the shoots were only 2.7-fold and 1.5-fold lower than those in T1_1 for the respective nutrients ($p \le 0.05$). In terms of micronutrients, including magnesium, sodium, and sulphur, T2_1 yielded higher contents than T1_1 ($p \le 0.05$), while the difference in calcium contents in T1_1 and T2_1 was insignificant (p > 0.05). It is noteworthy that the effect of magnesium addition in the fortified urine treatment (T3 1) hardly affected its contents in parsley compared to $T2_1$ (p > 0.05), suggesting that the formulation in UrVal fertiliser (T2_1) adequately addressed parsley nutrition needs.

3.2. Case II - rhipsalis growth under controlled glasshouse conditions

In this trial, the performance of UrVal fertiliser (T2_2) was compared against Ca-Mg grower (T1_2). Fig. 4 provides a comprehensive overview of the trial results, detailing the fresh and dry weights of rhipsalis shoots and roots, roots-to-shoots ratio, and stem length of rhipsalis throughout the experimental duration. Notably, the fresh biomass yield of both shoots and roots was observed to be higher under the T1_2 treatment compared to T2_2, resulting in 29.5 g and 24.0 g for shoots, and 2.2 g and 1.6 g for roots, respectively, depending on the respective treatments. On the other hand, the differences in dry weights of both shoots and roots between the treatments were negligible. Despite the differences in biomass yield, the roots-to-shoots average ratio remained relatively consistent at 0.8 across both treatments, suggesting a balanced growth pattern irrespective of the fertiliser type (p > 0.05). Regarding stem length, both T1_2 and T2_2 exhibited a progressive increase over the entire observation period. However, T1_2 consistently maintained a slightly longer stem length compared to T2_2, with stem lengths reaching up to 17 cm and 15 cm, respectively. It is worth noting that while the stems in the T1_2 group exhibited growth from 13 cm to 17 cm, those in the T2_2 group displayed slightly more modest growth, expanding from 10 cm to 15 cm.

Fig. 5 represents the tissue analysis results of rhipsalis shoots and roots under different fertiliser treatments. The total nitrogen and potassium contents in shoots and roots exhibited minor variations in their mean values between the treatments, however, these differences were statistically insignificant (p > 0.05). Consequently, all three major



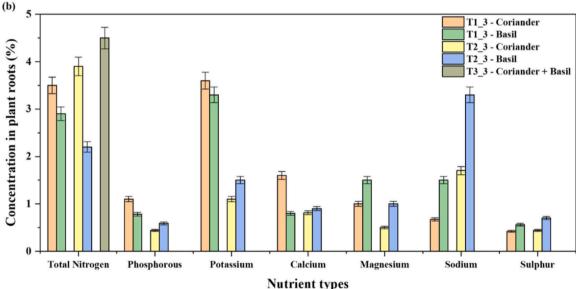


Fig. 8. Plant tissue analysis in coriander and basil (a) shoots; and (b) roots under different fertiliser types.

nutrient contents were nearly identical in T1_2 and T2_2 across both shoots and roots. Despite the urine fertiliser containing only half the concentration of phosphorous and potassium compared to the commercial fertiliser, their concentrations in the shoots and roots tissues were indistinguishable across the two treatments. Conversely, the Ca-Mg grower treatment (T1_2) yielded higher concentrations of calcium and magnesium in the shoots, with slight but statistically significant increases of 0.4% and 0.2%, respectively, compared to those observed in shoots treated with urine fertiliser ($p \le 0.05$). Interestingly, similar to the major nutrients, other micronutrient levels in the roots exhibited almost identical results across both fertiliser treatments (p > 0.05), suggesting a consistent uptake of nutrients irrespective of the fertiliser type employed. Overall, rhipsalis grown with urine fertiliser showed highly comparable results to the commercial fertiliser, indicating that the nutrient composition in urine fertiliser (T2_2) was sufficient for nutritional requirements of rhipsalis.

3.3. Case III – coriander and Basil growth in commercial indoor hydroponic systems

This case study aimed to investigate the efficiency of UrVal fertiliser (T2_3) in its application to a compact DWC hydroponic system

compared to the commercial fertiliser (T1_3). Furthermore, the inclusion of UrVal fertiliser with a high nitrite concentration (T3_3) highlights the persisting challenge of nitrite accumulation during the MBR process, which inhibits both ammonia-oxidising bacteria (AOB) and nitrite-oxidising bacteria (NOB) as well as requires additional chemicals to restore the balanced AOB/NOB activity and stabilise the process [13]. Nitrite is recognised for its detrimental effects on numerous plant species, thereby reinforcing caution against its incorporation in fertilisers [40]. However, as noted in the introduction, few studies have investigated the impacts of nitrite content in hydroponic treatment solutions. As such, evaluating of the impact of T3_3 in this trial can be crucial for identifying effective strategies to address the aforementioned challenge in MBR process.

Fig. 6 displays the fresh and dry weights of coriander and basil shoots and roots, along with the roots-to-shoots ratio under various fertiliser treatments. Basil biomass using urine fertiliser (T2_3) was significantly lower compared to the commercial fertiliser (T1_3), with yields of 42.5 g and 90.7 g, respectively ($p \le 0.05$). Similarly, coriander yield with urine treatment was approximately a third less than with commercial treatment, with yields of 42.4 g and 66.8 g, respectively ($p \le 0.05$). However, the roots-to-shoots ratio of coriander remained consistent at 0.1 across both treatment types (T1_3 and T2_3), while

that of basil showed slight but statistically significant variations, with ratios of 0.13 and 0.1 for T1_3 and T2_3, respectively ($p \le 0.05$). Fig. 7 depicts the stem lengths of coriander and basil, showing no statistically significant difference between the two treatments (p > 0.05), with lengths ranging from 29–30 cm for coriander and 26–27 cm for basil, although basil exhibited higher variability. Overall, urine fertiliser proved to be more effective in promoting coriander growth than basil, considering its more comparable results to commercial fertiliser.

In the case of treatments containing high nitrite (T3_3), basil growth response was significantly impacted, resulting in biomass yields of 1.1 g for basil and 27.3 g for coriander. The roots-to-shoots ratios of coriander and basil were notably lower in treatments with high nitrite compared to other treatments without nitrite. The toxic effect of high nitrite in hydroponic solution was evident in the stem length results, with lengths of 18.4 cm for coriander and 2.2 cm for basil. Furthermore, both plants exhibited weak and stunted growth, with leaves turning yellow due to nitrogen deficiency [41]. This indicates that nitrite nitrogen concentrations of 120 mg/L in the treatment solution significantly affected plant growth due to its toxic effect. However, a previous study reported that nitrite under 100 mg/L has negligible effect on the plant [42]. Thus, further studies are needed to investigate the threshold of nitrite concentration that can cause stunted plant growth.

Fig. 8 illustrates the concentration of each nutrient in the shoots and roots of both plants under different treatment solutions. Due to the lack of dried biomass for sample analysis in the case of T3_3, coriander and basil shoots were combined and described as T3_3 in Fig. 8a. Additionally, although the coriander and basil roots were combined, the sample amount was insufficient, thus only the total nitrogen content was measured, as described in Fig. 8b. The difference in total nitrogen content in shoots between T1_3 and T2_3 was marginal for both plants, ranging from 5.1--5.6% in coriander and 4.2--5.1% in basil. Despite the low biomass yield in T3_3, the total nitrogen content in shoots was highest at 5.7%. Notably, the largest difference was observed in nitrate concentration in the shoots. Shoots of T1_3-treated plants contained 1.8-2% nitrate, while those of T2_3-treated plants contained 0.4-0.6%. This difference can be attributed to the higher nitrate content ratio in the total nitrogen of the commercial fertiliser formulation compared to that in the urine fertiliser formulation. This finding also explains the higher yield of basil roots, as previous studies have reported that a high nitrate presence contributes to the growth of basil roots [43]. Conversely, the diminished nitrate content in T3_3 resulted in a reduced nitrate composition in the shoots, measuring at 0.15%. However, it is important to note that consuming leafy vegetables with significantly high nitrate levels and its accumulation in human body can pose health risks, such as methemoglobinemia or blue baby syndrome, which are related to blood oxygen levels [44-46]. The acceptable daily nitrate intake for humans set by the World Health Organisation (WHO) is 3.7 mg/kg bodyweight [47]. Hence, UrVal fertiliser may be a favourable option for the growth of hydroponic plants, considering its comparable yield to commercial fertiliser and the potential reduction of nitrate accumulation risk in human body.

4. Conclusion

In this study, the efficacy of urine-derived UrVal fertiliser, produced through biological nitrification and concentration via distillation, was evaluated under different experimental conditions. The results indicated that the UrVal fertiliser showed comparable performance to commercial fertilisers in cultivating parsley, rhipsalis, coriander, and basil, regardless of the growth system. Additionally, the micronutrients supplementation on the UrVal fertiliser yielded negligible improvements in plant growth response. Notably, the high nitrite content in UrVal fertiliser resulted in inhibited growth, due to the toxic effect of nitrite. This finding highlights the need for further research to establish the threshold nitrite concentrations that cause toxicity, addressing a critical challenge in the urine treatment MBR process. Continued

research is also necessary to optimise nutrient formulations for different plant species and growth conditions to ensure optimal growth and yield. In conclusion, urine-derived fertilisers hold significant promise as a sustainable nutrient source for hydroponic growth, aligning with principles of circular economy. However, the broader application of anthroponics in urban settings faces several challenges, including process scalability and optimisation, economic viability, policy and regulatory development, and public acceptance. A notable challenge is the need for additional plumbing infrastructure for urine diversion and collection in existing urban buildings. Therefore, extensive and multidisciplinary research is essential to develop a comprehensive pathway for integrating anthroponic systems into urban agriculture initiatives, ultimately contributing to a more sustainable and resilient nutrient recovery system.

CRediT authorship contribution statement

Stefano Freguia: Writing - review & editing, Project administration, Investigation. Sherub Phuntsho: Writing - review & editing, Supervision, Project administration, Funding Conceptualization. Li Gao: Funding acquisition. Jing Guan: Writing review & editing. Sungyun Lee: Writing - review & editing, Investigation. Hokyong Shon: Writing - review & editing, Writing original draft, Visualization, Validation, Supervision, Resources, Project administration, Methodology, Investigation, Funding acquisition, Formal analysis, Data curation, Conceptualization. Weonjung Sohn: Writing - original draft, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. Andrea Merenda: Writing - review & editing, Investigation, Formal analysis. Ibrahim El Saliby: Writing review & editing, Methodology, Formal analysis.

Data Availability

No data was used for the research described in the article.

Declaration of Competing Interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: 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. Ho Kyong Shon serves as a co-Executive Editor for the DWT journal, while the editorial handling and review of this manuscript were overseen by a different co-Executive Editor.

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Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.dwt.2024.100682.

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