



Review Article



Source-Separated Human Urine as Fuel for Microbial Fuel Cell: A Mini Review

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Abstract

Source separation and treatment of human urine has drawn much attention in the past 20 years as an environmentally friendly approach for recovering nutrients and energy. However, the majority of existing urine treatment technologies including ammonia stripping, freezing-thawing, and nanofiltration require much energy input. These technologies add significantly more to humanity's environmental footprint than microbiological technologies. This paper reviews the existing Microbial Fuel Cell (MFC) for recovering energy and nutrients from source-separated human urine. Experimental studies in this area are still in their early stages of development and more researches considering the unique property of urine instead of MFC system optimization should be done in the future. In particular, evaluation and control of effluent quality, urine hydrolysis with the existing of bacteria in MFCs, efficiency of nutrient and energy recovery, the impacts and removal of heavy metals and micropollutants deserve study.

Keywords

Microbial Fuel Cell; Nutrients Recovery; Source-Separated Human Urine

Introduction

Humanity's environmental footprint, especially its carbon, material, and water footprints, has already far exceeded the maximum sustainable level as a result of the rapid increase in population and expansion of technology in the past 20 years [1]. As such, it is becoming a common practice to recover energy and nutrients from waste resources and thus reduce the environmental impacts of anthropogenic activities to more sustainable levels [2-4]. Domestic wastewater, as the main waste product of human activities, is rich in nutrients. However, both its treatment and discharge add substantially to the carbon, material, and water footprints. Hence, more environmentally friendly wastewater treatment approaches and technologies have been proposed for recovering energy and nutrients and for treating the water for subsequent reuse [2,5,6]. Source separation or decentralized treatment for special waste streams are presently the most efficient approaches to recover nutrient and energy [6-8]. Urine contributes 7-14% of Chemical Oxygen Demand (COD), 69-80% of Nitrogen (N), 40-50% of Phosphorus (P), and 60% of Potassium (K) in domestic wastewater yet it only represents 1.3-1.6% of its total volume [9,10]. Thus, it is an ideal waste stream that can be separated at the source to recover nutrients and energy.

The concept of separating urine at the source and then treating it is not new [10-12], and practical engineering approaches for urine separation, which

can achieve 70-75% urine recovery, have been designed and applied since the late 1990s [13-16]. However, the research in this area only began in earnest after the modeling results confirmed the benefits of separating urine at the source, viz. reduced energy consumption and extended lifespan of the wastewater treatment plant [17-19]. Later, various urine treatment techniques were investigated, as comprehensively reviewed by Maurer et al., [20]. However, most of these techniques focus on stabilizing urine and removing nutrients instead of recovering nutrients and energy. Currently, the two main approaches for recovering nutrients from urine are 1) chemical dosing for P and partial N recovery as struvite [21,22], and 2) zeolite adsorption or air stripping for N recovery [23-27]. Both approaches require chemical or mineral addition, which results in a higher material footprint. According to recent Life Cycle Assessments (LCAs) and comparisons of management approaches for separating urine at the source, it is still environmentally acceptable if only small amounts of chemicals are used and the efficiency of nutrient recovery is sufficiently high [28,29]. Hence studies have been conducted on the use of seawater [30,31], bitter [32], and wood ash [33] as precipitant aids to recover P and N from urine as struvite, which can easily achieve an acceptable P recovery efficiency (>90%) [21,22,30,32]. However, the recovery efficiencies for other nutrients, i.e., N and K, are still very low.

Considering that the composition of urine and the nutrient requirements of most crops and plants are similar [34,35], it is desirable to directly apply urine in agriculture to achieve a higher nutrient application efficiency [36-38]. However, a significant amount of research has shown that the microbiology in human urine, its high salinity, and its strong concentrations of ammonia, heavy metals, and micropollutants are all detrimental to the growth of plants, and the conditions of the soil and the ecosystem [11,12,39-42]. Hence, other environmentally friendly methods that can yield high efficiencies for nutrient recovery but low impurities in the final products are needed. On the other hand, the use of concentrated organic matter in source-separated urine for energy production should also be considered. Microbial Fuel Cell (MFC) offers a sustainable way to recover energy and nutrients from wastes and have been widely studied and applied. However, its application in urine treatment is still in its infant stages. To explore the feasibility of using human urine as the fuel for MFCs, this study summarizes the composition of human urine reported in published studies to re-evaluate its potential for treatment with MFCs for subsequent nutrient and energy recovery. Moreover, the existing literature on treating source-separated urine with MFCs is comprehensively reviewed to better understand the research status. The recommendations for future studies on using MFC for separated urine treatment are given based on the literature review.

Characterization of Human Urine

The properties of human urine have been well documented in the past, mostly for human health monitoring but also for urine treatment. The characteristics of urine are largely affected by factors such as dilution or storage time, and any pre-treatment will further affect its composition and the performance of the subsequent treatment processes. Urine excreted by a healthy human is mildly acidic with high conductivity because of a high ion concentration. Besides plenty of ions, urine also contains a large number of organic compounds mainly made up of carbon and nitrogen. Urea, as an essential component of urine, contributes around 80% of Total Nitrogen (TN) in fresh urine. Its concentration can vary from 6000 to 23300 mg/L. Therefore, urea is the primary compound to be considered for nitrogen recovery. The Total Organic Carbon (TOC) concentration in urine can reach 8000 mg/L. Such a high concentration indicates that urine is also an ideal waste stream for energy recovery although the high TOC concentration is largely reduced after urine hydrolysis [30]. Even though the ionic strength of urine is very high, the major contributions come from 10 ions: Ammonium (NH_4^+), Sodium (Na^+), Potassium (K^+), Calcium (Ca^{2+}), Magnesium (Mg^{2+}), Chlorine (Cl^-), Phosphate (PO_4^{3-}), Carbonate (CO_3^{2-}), Bicarbonate (HCO_3^-), and Sulphate (SO_4^{2-}). All the ion concentrations in urine vary within large ranges. NH_4^+ and inorganic-C concentrations will increase with urine hydrolysis, while Ca^{2+} , Mg^{2+} , and PO_4^{3-} will decrease due to precipitation.

Upon coming into contact with the natural environment, urea hydrolyse to ammonia in a process known as urine hydrolysis, producing ammonia, bicarbonate and resulting in a high pH. The biochemical reaction is as follows: $(\text{NH}_2)_2\text{CO} + 2\text{H}_2\text{O} \rightarrow \text{NH}_3 + \text{NH}_4^+ + \text{HCO}_3^-$ [21]. Urine hydrolysis is mediated by the enzyme urease, which is ubiquitous in the environment. Urease can be produced by bacteria, algae, and plants. The presence of urease makes the urine hydrolysis rate 1013 times higher than uncatalyzed urine hydrolysis [43-46]. This indicates that the urine hydrolysis rate and extent can be manipulated to meet the pH and ammonia concentration requirements for urine treatment. To understand the urine hydrolysis process and functional organisms involved, four studies that compare fresh and hydrolysed urine under different conditions are shown in table 1 [21,30,46,47]. Previous research has studied the occurrence of urine hydrolysis in the presence of either indigenous bacteria in urine or seeding sludge in the natural environment (Table 1), urine hydrolysis at different temperatures (2.5-30°C) and percentages of hydrolysis urine addition as seeding [48]. The required duration for complete hydrolysis may vary from less than 20 minutes to 1 month (Table 1). After urine hydrolysis has taken place, the pH increases from around 6 to 9, conductivity increases nearly

Parameter	Udert et al., [21]		Dai et al., [30]		You et al., [47]		Tang et al., [46]	
Treatment	Stirring in flasks		Storage in sealed container		Microbial Fuel Cell (MFC)		SUPR reactor ³	
Duration	4.5 hours		1 month		16 minutes		6 hours	
Dilution solution	No		No		No		Seawater	
Dilution ratio	No		No		No		1:1 dilution	
Seeding	Solids in No Mix pipes		No		Activated sewage sludge		Seawater and fresh urine	
Parameter	Fresh	Hydrolyzed	Fresh	Hydrolyzed	Fresh	Hydrolyzed	Fresh	Hydrolyzed ⁴
pH	6.0 ± 0.3	8.7 - 8.9	6.5 ± 0.3	9.2 ± 0.1	6.57	9.24 - 9.28	6.67	9.31
Conductivity (mS/cm)			14 ± 4.5	39 ± 2.2	12.3	22.5 - 23		
TOC (mg/L) ¹	3638 ± 2		6100 ± 1900	2500 ± 250	2720	2550		
NH ₄ ⁺ (mg N/L)	386 ± 3	1200 - 2200	330 ± 75	6400 ± 970	363 ± 8	3231 - 3278	261	2793
Ca ²⁺ (mg/L)	168 ± 2	50 (30%) ²	150 ± 45	14 ± 12 (9.3%) ²				
Mg ²⁺ (mg/L)	121 ± 2	1.2 (1%) ²	36 ± 9.1	0.8 ± 0.5 (2%) ²				
PO ₄ ³⁻ (mg P/L)	559 ± 5	313 (56%) ²	350 ± 150	250 ± 85 (71%) ²	202 ± 6	238 - 265	178	4.99
Inorganic-C (mg C/L)	< 5		56 ± 28	3500 ± 510				

Table 1: Composition and physicochemical properties of hydrolyzed urine with different treatment methods at the room temperature of approximately 20°C (the parameters differ from those of fresh urine).

¹TOC is calculated from total COD, if available, for comparison.

²Value (percentage left in urine after urine hydrolysis).

³Seawater-based urine phosphorus recovery reactor.

⁴Results with a 1:1 ratio of seawater dilution.

twofold due to the release of inorganic carbon, and high concentrations of ammonia of between 1200-6400 mg NH₃-N/L are produced. These can negatively impact on the urine treatment efficiency as most of the organisms are sensitive to the high pH and high concentrations of ammonia [49,50]. Furthermore, the precipitation of P as struvite, triggered by the high pH after urine hydrolysis, leads to just 1-2% of the Magnesium (Mg) in fresh urine remaining in the hydrolysed urine. Moreover, the loss of P can exceed 40% if the cation (mainly Mg and Ca) concentration in fresh urine is sufficiently high for P precipitation. The excessive loss of P and Mg may result in low P recovery and insufficient Mg when urine is used as the sole nutrient source.

Besides the parameters mentioned above, heavy metals [11,12] and micropollutants including estrogens, androgens, pathogens, and pharmaceuticals [39,40,51-53] are also present in human urine. Their removal in the urine treatment processes should also be investigated to mitigate their negative impacts on the ecosystem and environment.

Energy and Nutrient Recovery Using MFCs

Working principle of MFC

MFC is a device utilizing bacteria as the catalysts to convert the energy of organic or inorganic matter to electricity. Similar

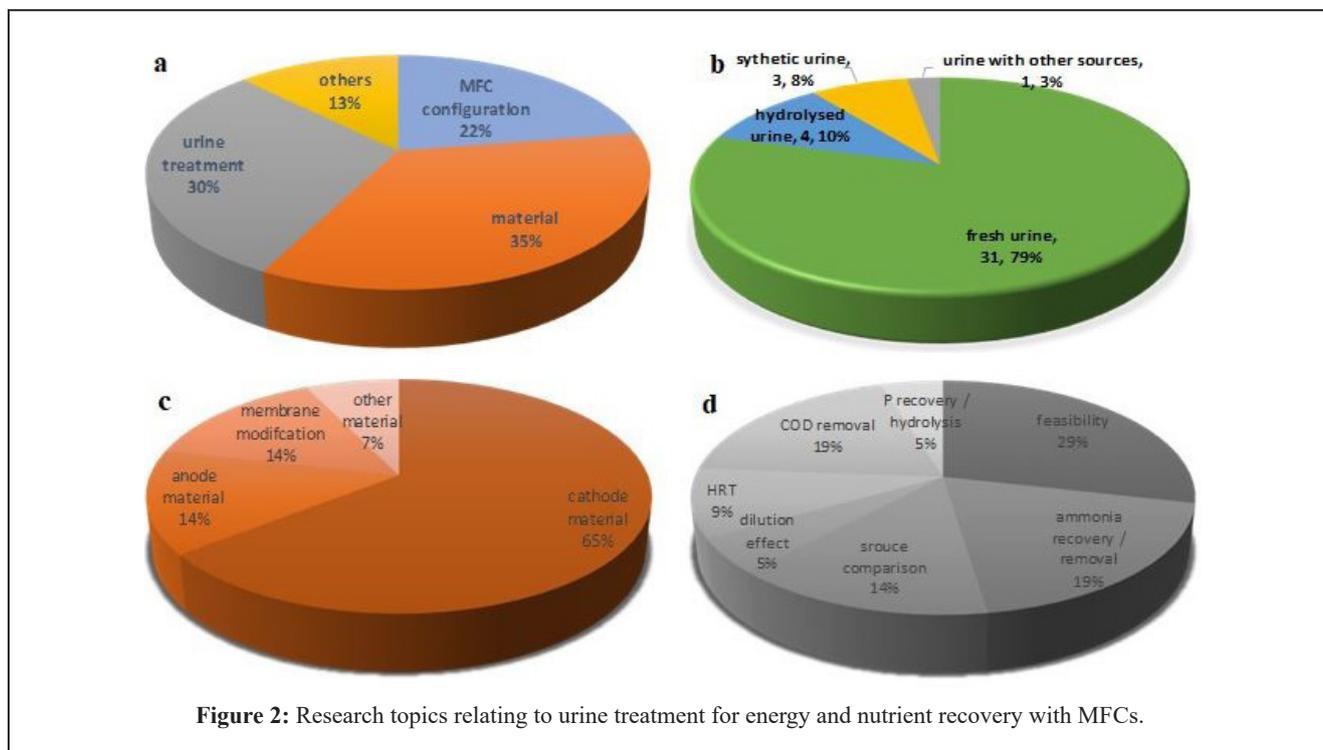
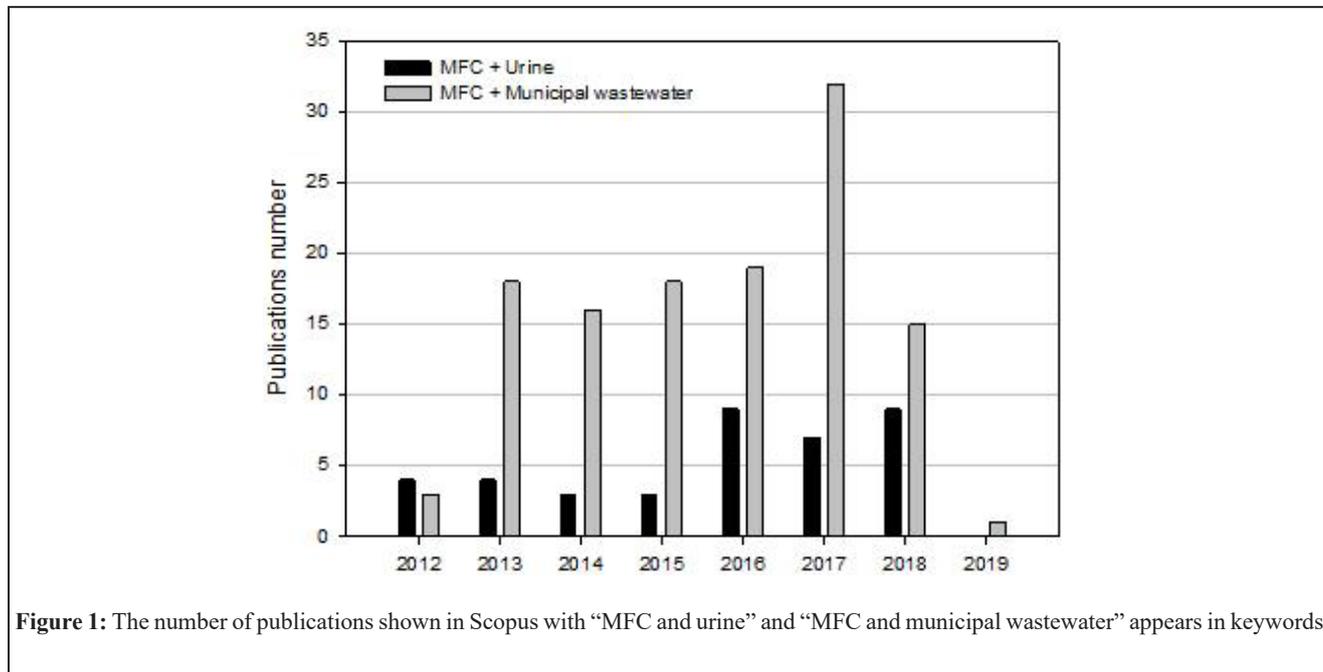
to conventional chemical fuel cell, MFC consists of electrolyte and two electrodes connected through an external circuit. On the anode, bacteria catalyze the oxidation of reduced substrates, releasing some of the electrons produced from cell respiration to this electrode. Therefore, fuel with high organic or inorganic matter to feed in anode is the intrinsic requirement for running an MFC. On the cathode, electrons transferred from external circuit reach this electrode and combine with protons that diffuse from anode to sustain the current generated [54]. Generally, oxygen is the terminal electron acceptor in MFCs. Electrons and protons react with oxygen on the cathode, aided by certain catalyst like platinum, to form water.

Literature benchmarking

An MFC is a bio-electrochemical system and is considered as the next generation of wastewater treatment technology for energy production, which was begin from the beginning of 2000. However, there are many challenges for the development of this technology to full-scale application [35,54-57]. Studies on MFCs operated with different substrates, especially real wastewater, show that the low conductivity and buffering capacity of wastewater are two drawbacks, which deteriorate the long-term performance of the MFC and thus limit its application [35,58,59]. However, source-separated urine represents an ideal substrate for MFCs given the natural high conductivity and carbonate-bicarbonate pH buffering system arising from the urine hydrolysis. Moreover, the high

substrate concentrations in urine can benefit the MFC [60]. Despite the high potential of using urine as fuel for MFCs, experimental studies on its feasibility began only recently. Literature benchmarking for this research area is conducted with Scopus and summarized in figure 1 and 2.

studies focused on MFC itself instead of urine treatment. More than 50% of studies mainly focused on the MFC configuration and electrode materials modification (Figure 2a). Although all these studies applied urine as the fuel feeding to the anode chamber, no study claimed that the material was specially



As indicated in figure 1, the study on treating urine with MFC was not extensively conducted yet comparing with the investigation of using wastewater as the fuel of MFC. Most of

designed for urine treatment optimization according to the unique property of urine. Only 30% of published studies investigated the impact parameters relating to urine treatment

(Figure 2a). The topic of urine treatment with MFC includes feasibility study, energy and resources recovery potential, pollutants removal efficiency, and optimum conditions (Figure 2d). Most of the studies used undiluted fresh human urine as the sole substrate for the MFC without pH control (Figure 2b). A few studies used ureolyzed human urine, synthetic urine, or urine with additional sea salts. Despite different MFC configurations with various materials were used as the anode and cathode in these studies, all of them confirmed the feasibility of using all types of urine as fuel to produce energy in the MFC. However, the electricity production of these MFCs fed with human urine is poor compared with that of MFCs using other substrates as fuel [56]. Therefore, the MFC materials and configuration optimization was the major research topic in this field. Considering unique property of urine, the operation condition optimization aiming at both nutrient and energy recovery should be another valuable research topic in urine-MFCs studies.

MFCs materials and configuration comparison

The most promising method to enhance the electricity production or coulombic efficiency is the material modification when same fuel was fed in the system. Therefore, more than half studies on MFCs fed with urine investigated the impacts of anode materials, cathode materials, membrane, supporting materials, and MFC configurations on the power density of MFCs. In MFCs, the electrogenesis flora growth on anode material surface is the intrinsic reason for power production, so the relationship between anode surface area / volume and power density obtained from majority of urine-MFCs studies were summarized in figure 3 according to wide literature review.

volume, because the data are extracted from different studies conducted in various MFCs with totally different operation conditions. However, it can be observed that the highest power densities are appeared when anode surface area lies from 300 to 600 cm^2 (Figure 3a), Moreover, the power density shows a reducing trend when the anode volume increases (Figure 3b), which is consistent with the major finding in previous studies [61,62].

The anode materials comparison was seldom investigated, which was mainly carbon fiber veil/brush/sleeve in most of studies [63,64]. Chen et al., [65] introduced super-capacitor activated-carbon and super-capacitor activated-carbon as double-layer or multiple-layer anode material and enhanced the power density 2.9 times [65]. Most of the material modification studies focus on cathode [66,67]. Platinum [68,69], graphite powder [70,71], polyurethane [72], egg [73], and Polytetrafluoroethylene (PTFE) [66,74,75] were used as the conductive paint on cathode material. Comparing with all types of cathode painting materials, 20% of PTFE had the best performance and gave a power density as high as 16 W/m^3 [66]. Gajda et al., applied iron and nicarbazin catalyst for breathing cathode in a MFC fed with fresh urine and the power curve showed a maximum power density of 48 W/m^3 [76], which is the highest value in the urine-MFCs till now. However, no material modification and optimization according to the unique properties of urine before and after hydrolysis was conducted, which is an interesting research topic to be explored in the future. In general, the MFC configurations modification and scale up were mainly conducted by Professor Ioannis Ieropoulos's research group. They kept trying to scale-up the urine-MFC system with different configurations in series or parallel connections [73,77-

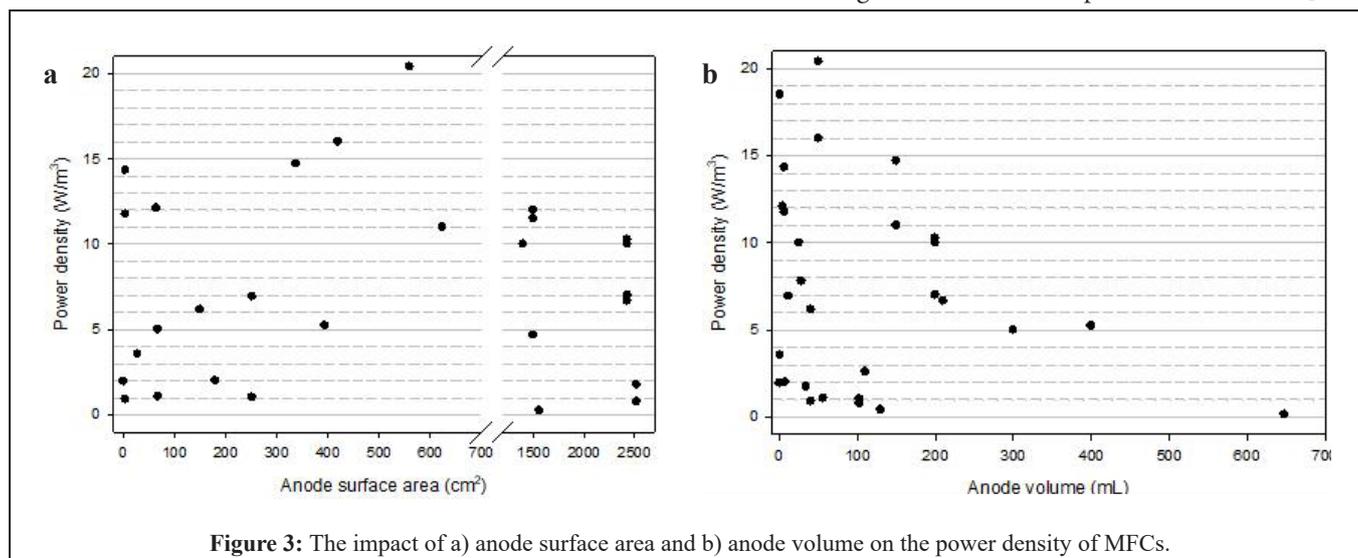
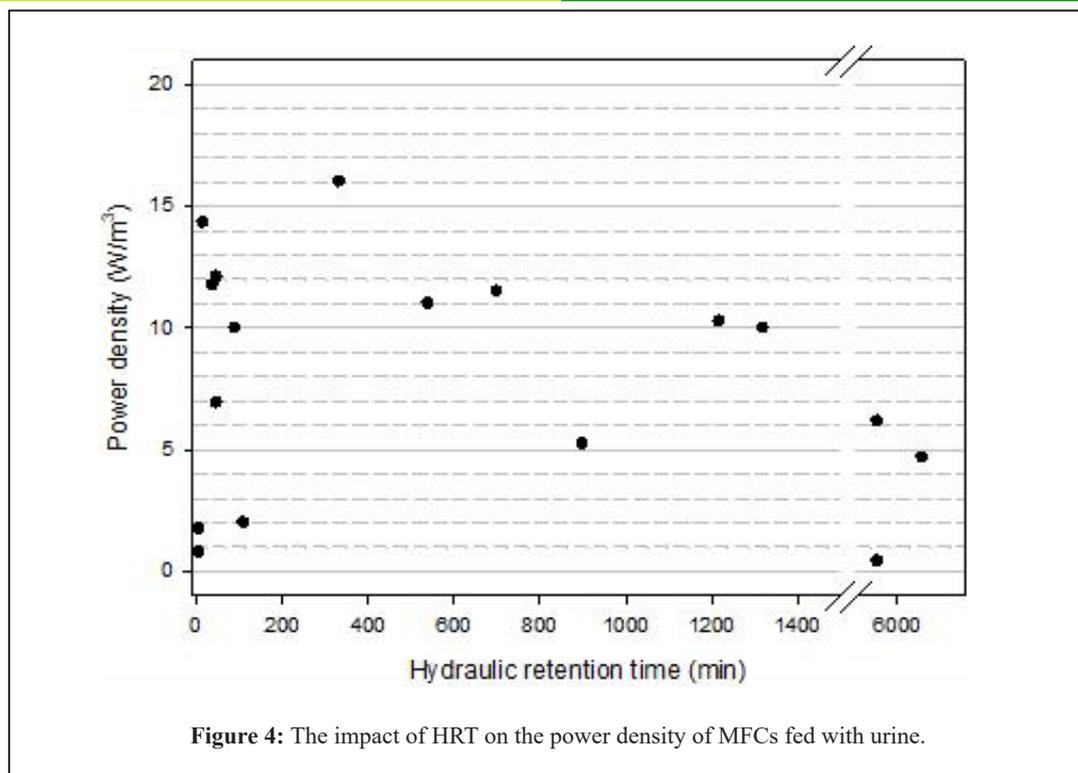


Figure 3: The impact of a) anode surface area and b) anode volume on the power density of MFCs.

As shown in figure 3, no statistical relationship can be clear drawn between anode surface area and MFCs. All these results are only attributed by the variation of anode surface area and

79]. However, the power density of large scale MFC fed with human urine is very low comparing with the small system. Therefore, more exploration on the MFC system scale up is necessary.



In general, the maximum power density decreases with the increase of HRT when HRT is longer than 200 minutes. However, the maximum power density of MFC systems varied in a very large range when HRT is lower than 100 minutes, because other parameters are more important than HRT when COD is excessive. Most of the above studies applied fresh urine as fuel, which have different COD concentrations. A previous study investigated the impact of COD concentration on power density of MFC fed with cow's urine with different dilution ratios, indicating that the optimum COD concentration was 3000 mg/L in the range from 1500 to 30000 mg/L at the same HRT [84]. It suggests that an optimum loading rate considering the impact from both HRT and COD concentration should be more reasonable.

The urine hydrolysis in MFCs is also lack of investigation. The characteristics of urine significantly changes after hydrolysis. Before hydrolysis, the anode reaction follows the equation: $\text{CO}(\text{NH}_2)_2 + \text{HOH} \rightarrow \text{N}_2 + \text{CO}_2 + 6\text{H}^+ + 6\text{e}^-$ which can oxidize organic nitrogen to nitrogen gas and further reduce the pH value at anode side [84]. If the hydrolyzed urine was fed in MFCs, the pH can be as high as 9 and no urea can be provided as carbon source for anode reaction. In this case, other complicated COD can be utilized by bacteria attached on the anode material of MFCs for energy production. Both fresh urine and hydrolysed urine have its strong advantages to be fuel for MFCs. Only one excellent study proved the possibility of fresh urine hydrolysis in MFC with short HRT, followed by phosphorus precipitation, and then COD degradation with

MFC for energy production [47]. However, no researcher compared the impact of urine hydrolysis in MFC system.

Nutrient recovery and reuse

High concentration of nitrogen and phosphorus is a prominent feature of urine. Both nitrogen and phosphorus are essential nutrients for living creatures. Therefore, nutrient recovery from urine is always regarded as a meaningful research and engineering topic. With regard to N recovery from urine, besides the zeolite adsorption or air stripping, the most-studied approach is the electrochemical extraction of ammonia coupled with ion exchange membranes. Examples of this approach include electrodialysis cells [85,86] and Microbial Electrolysis Cells (MECs) [87,88]. For complete recovery of N, P, and K from urine, the most-studied methods are nitrification followed by distillation [89], nanofiltration for removing micropollutants and purifying nutrients [52], and freezing-thawing for concentrating nutrients [90]. Apart from MFCs, all the methods require an extensive energy input with inevitable N loss. However, the transport or oxidation of ammonia in the MFC fed with human urine, the impact of high ammonia concentrations on the electricigens, and the potential for ammonia recovery are still unknown. Two studies confirmed the absence of nitrifiers or anaerobic ammonia-oxidizing bacteria in the MFC fed with human urine [91,92], indicating the possibility for ammonia recovery in such system. Despite Kuntke et al., [64] and Zhou et al., [92] achieving 50% of ammonia recovery from urine, the recovery efficiency should

be optimized further.

Magnesium or calcium dosage for phosphorus recovery from urine is a widely accepted approach. The cation source can be replaced by seawater due to its high magnesium concentration [30]. The cation dosage will be more effective after urine hydrolysis because high pH value and excessive ammonia can be provided after hydrolysis to facilitate the precipitation process, especially for struvite formation. Despite a few studies confirming the occurrence of urine hydrolysis in the MFC when fresh urine was fed into the system [47,68,69], the rate of hydrolysis and the long-term stability of power generation have not yet been evaluated. Seawater or sea salt was also considered to be fed in MFC with fresh urine for simultaneous phosphorus recovery and energy production in one study. The results show that the addition of sea salt can realize phosphorus recovery and enhance the energy production [74]. Beside nitrogen and phosphorus, several studies also reported that heavy metals can be recovered from urine from catholyte of MFC [93,75], which can be a good research topic.

Outlook

More than 80% of the research works relating to the urine-MFC have been conducted by a research team in United Kingdom, which is led by Professor Ioannis Ieropoulos since 2012. This makes the research direction of using urine as fuel of MFC is lack of diversity. Research directions relating to the unique properties of urine should be extensively explored, which are listed out as following.

- To focus more on nutrient recovery from human urine with MFCs instead of power generation.
- To investigate the nutrients transformation in urine in the MFC systems.
- To explore the urine hydrolysis in the MFCs.
- The study the conversion or removal of urine-derived COD, phosphate, and sulphate in the MFCs.
- To analyze the impacts of complex compounds in human urine on the performance of the MFCs.
- To extensively investigate the removal of heavy metals and micropollutants and their impacts on the bacteria in the MFCs.
- To study the impacts of loading rate on the performance of MFCs.

From the existing literature on the topic of treating human urine using MFCs, energy production appears to be the second goal after nutrient recovery. In future studies however, quick urine hydrolysis, ammonia recovery, and the removal of micropollutants and their impacts should become the focus. After extensively understanding the mechanisms governing the

treatment of human urine using MFCs, the system scale-up should be pursued to realize the practical application of this technology.

Conclusion

The treatment of source-separated urine for recovering energy and nutrients has the potential to reduce humanity's footprint on the environment to sustainable levels and is necessary for future sustainability. Studies on the MFCs for separating human urine at the source are only just starting. More experimental research in this field must be performed to evaluate and optimize the existing technologies.

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