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Use of Tangerine Waste as Fuel for the Generation of Electric Current

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Abstract: Fruit waste has increased exponentially worldwide, within which tangerine is one of those that generates a greater amount of organic waste, which is currently not fully used. On the other hand, microbial fuel cells (MFCs) are presented as an opportunity to take advantage of organic waste to generate electricity, which is why the main objective of this research is to generate bioelectricity using tangerine waste as a substrate in microbial fuel cells using zinc and copper electrodes. It was possible to generate current and voltage peaks of 1.43973 ± 0.05568 mA and 1.191 ± 0.035 V on days eighteen and seventeen, respectively, operating with an optimum pH of 4.78 ± 0.46 and with electrical conductivity of the substrate of 140.07 ± 3.51 mS/cm, while the Brix degrees gradually decreased until the last day. The internal resistance determined was 65.378 ± 1.967 Ω, while the maximum power density was 475.32 ± 24.56 mW/cm^2 at a current density of 5.539 A/cm^2 with a peak voltage of 1024.12 ± 25.16 mV. The bacterium (*Serratia fonticola*) and yeasts (*Rhodotorula mucilaginosa*) were identified in the substrate with an identity of 99.57 and 99.50%, respectively. Finally, the cells were connected in series, managing to generate 3.15 V, which allowed the turning on of a red LED light.

Keywords: microbial fuel cell; bacteria; yeast; bioelectricity; tangerine waste

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1. Introduction

Fruits are of great importance in the diet of any human being, and in recent years the consumption of various types of fruits has begun to increase exponentially due to their various properties such as high content of bioactive components (including antioxidants), pigments, flavor compounds, proteins, essential oils, enzymes and dietary fibers [1–3]. It has been reported that approximately 124.73 million metric tons (MMT) of citrus, 114.08 MMT of bananas, 84.63 MMT of apples, 74.49 MMT of grapes, 45.22 MMT of mangoes and 25.43 MMT of pineapples have been produced in 2018; an increase of 60% is estimated for the year 2025 [4,5]. Due to this, the waste of fruits would also increase, which would generate losses for the companies and farmers dedicated to harvesting, buying and selling it [6]. In the year 2020, the European Union estimated that about 89 million tons of waste from different types of food (vegetables and fruits) were generated, and an annual loss of fruits and vegetables of 21 million tons was estimated, which would represent an approximate loss of 10.6 billion dollars [7,8]. The countries with the greatest development worldwide apply different types of methods for the decomposition and reuse of organic waste. Among the most important is the drying method, which consists of four steps: bisecting, biostabilization, solar drying and thermal drying [9,10]. Thus, there are also other methods (biochemicals recovery, vermicomposting, composting, etc.) for the treatment of fruit and vegetable waste. Essentially, all of them have the same process, which is the collection of waste to a collection center where the most appropriate method is used depending on the material to be treated, which can be to produce biogas, leachate, bioelectricity, etc. [11–13].

One of the most consumed products are citrus fruits, where 18% of the world production of this type of fruit has an industrial use, such as the manufacture of juices, bioactive essential oils, jams, etc. [14,15]. It has been reported that the volume of citrus processed each year is 31.2 million tons. This in turn generates large amounts of waste (peel, pulp and seed residues), generating a high economic and environmental cost for the management of this residue amount [16,17]. The drawbacks for proper management of these residues are exacerbated in developing countries. Citrus ferment easily because they are highly biodegradable, chemically complex and bulky [18]. Within the citrus is the orange, which is a highly consumed fruit worldwide (with almost 10 million tons each year), with its highest consumption in juices; orange juice generates almost 50% of the mass of the fresh fruit. The countries that have a high percentage of orange production are China (28.16%), Brazil (12.24%), Mexico (5.60%) and India (8.82%), while those that import the most are Russia (10.63%), Germany (7.54%), France (7.19%), USA (6.44%) and Netherlands (6.72%) [19–21].

On the other hand, microbial fuel cells (MFCs) show promise as a technology for companies and farmers because they can generate bioelectricity from different types of substrates (wastewater, food waste, fruit waste, residual sludge, etc.), using them as fuels [22]. This technology has different types of design, but it basically consists of two chambers (anodic and cathodic) almost always joined by a proton exchange membrane inside, where the electrodes (anodic and cathodic) that are inside the chambers meet. They are joined on the outside by an external circuit [23,24]. MFCs use chemical energy to convert it into electrical energy, mainly due to the oxidation and reduction processes that occur [25]. Fruit waste in MFCs has not yet been studied in depth, and many types of fruit have not yet been reported by the different groups of researchers. However, some already exist, such as that of Rincón et al. (2022), who managed to generate approximately 300 mV and 41.3 mW/m^2 of voltage and power density, respectively, in their single-chamber MFCs using banana debris as a substrate [26]. Likewise, tomato waste has also been used in single-chamber MFCs, managing to generate peaks of 4 A and 4.2 V of electrical current and voltage; these high values may be due to the metallic electrodes (zinc and copper) and the volume (20 Kg) used [27]. Golden berry debris in single-chamber MFCs has also been reported, managing to generate peaks of 1.03 ± 0.02 V and 4.945 ± 0.150 mA, with an internal resistance of $194.04 \pm 0.0471\ \Omega$ [28]. It has been observed in the literature that high values of electric current, voltage and power density have been obtained using metallic electrodes and that the majority of waste used is wastewater and sludge, while fruit waste has not yet been widely addressed. However, it has been shown that the electrical performance of an MFC depends on the exoelectrogenic bacteria, and these in turn depend on the pH values at which they will operate in the MFC [29], as well as the durability of electricity generation of the type of electrode device used (anodic and cathodic) and biofouling in proton exchange membranes [30,31].

This research has the main objective of generating bioelectricity through single-chamber microbial fuel cells on a single-chamber laboratory scale, using tangerine waste as a substrate (fuel) and using zinc and copper metal electrodes. For this, the values of voltage, electric current, pH, electrical conductivity and Brix degrees are monitored; likewise, the internal resistance of the MFCs and the generation of their power density and electrical current density were measured. Likewise, microorganisms adhering to the anode electrode of the MFCs were molecularly identified. This research will give great contributions to generate electricity in a sustainable way for companies and farmers, where they can use the waste from their own products for electricity.

2. Materials and Methods

2.1. Fabrication of Microbial Fuel Cells

The MFCs used were purchased from SAIDKOCC Manufacturing (SAIDKOCC-10091720, Fujian, China), where the copper anode (Cu, with an area of 40 cm^2) and zinc cathode (Zn, with an area of 62.5 cm^2) electrodes were placed inside and outside (one side of the electrode in contact with the environment) of the cell. The electrodes were connected by an external circuit that consisted of a 6 mm copper wire and a 100 Ω resistance (three MFCs were used). The anode and cathode chambers were separated by a proton exchange membrane (PEM-Nafion 117; Wilmington, NE, USA), which was attached to the cathode electrode (in total, three MFCs were used); see Figure 1.

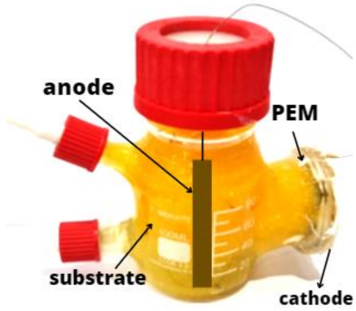

Figure 1. Schematization of the design of the MFC.

2.2. Obtaining Tangerine Residues

The tangerine residues were selected by the merchants of the Mercado La Hermelinda, Trujillo, Peru; they managed to collect 1.5 Kg. The collected residues were washed with distilled water (3 times) to eliminate any type of environmental impurities, and later let dry at room temperature (21 ± 1.5 °C) for 24 h. The tangerine waste passed through an extractor (Labtron, LDO-B10-Camberley, UK), able to obtain juice from the waste to the total of 650 mL.

2.3. Characterization of Microbial Fuel Cells

To monitor the electrical current and voltage parameters, a multimeter (Testech, KT-5510) was used. For the measurement of current density (DC) and power density (PD), the formula described by Rojas et al. (2022) was used, where DC = I/A and DP = IV/A (where V is the voltage and A is the area) [32], and where I is the current generated using the following external resistors: 1.5 ± 0.2, 5 ± 0.3, 10 ± 0.1, 20 ±2, 50 ± 4.2, 95 ± 8.3, 210 ± 15, 500 ± 22.4, 768 ± 23 and 995 ± 25 Ω. The values of electrical conductivity (conductivity meter- CD-4301), pH (pH meter/110 Series Oakton) and ° Brix (RHB-32 brix refractometer) were monitored for 30 days.

2.4. Isolation of Microorganisms from the Anode

To identify possible electrogenic microorganisms, a swab of the anode surface (with evidence of microbial growth) was performed. For the isolation of bacteria, nutrient agar and MacConkey agar were used, which were incubated at 30 °C for 24 h. For the isolation of fungi, Sabouraud agar was used and incubated at 30 °C for 24 h. To observe the microscopic characteristics, a Gram stain (for bacteria) and a lactophenol stain (for fungi) were performed. Finally, pure cultures of the isolates were made on inclined agar, for subsequent molecular identification.

2.5. Molecular Identification

Axenic cultures were sent to the BIODES laboratory (Laboratory of Integral Solutions Limited Liability Company, MI, USA) for molecular identification. Genomic DNA extraction and PCR amplification were steps prior to sequencing through the Sanger method.

Subsequently, the sequences were analyzed in the bioinformatics program MEGA X and then aligned in the BLAST to obtain the percentages of the identity of each isolate.

3. Results and Analysis

In Figure 2a, the voltage values monitored during the 30 days are shown, and it can be observed that the values increase from the first day (0.204 ± 0.005 V) until the seventeenth day (1.191 ± 0.035 V) and then slowly decline to 0.72623 ± 0.023 V in the last day. According to Kebaili et al. (2020) the oxidation and reduction reactions that occur inside the cells are responsible for the initial voltage values, because they quickly converts chemical energy into electrical energy [33]. The high voltage values observed, as discovered by Liu et al. (2023), would be due to the natural polyphenols derived from the fruits because they improve the biodiversity and abundance of electron-producing bacteria, which improve the overall performance of the system [34]. The voltage values shown are higher than those reported by Latif et al. (2020), where they used fruit waste (oranges, pineapples, bananas, papaya and mango) in their laboratory-scale MFCs with carbon felt electrodes and managed to generate maximum voltage peaks of 800 mV in the orange waste [35]. In Figure 2b, it is possible to observe the values of the electric current generated during the monitoring, where the values increase from 0.15959 ± 0.04933 mA (on the first day) to 1.43973 ± 0.05568 mA (on day 18), after falling by the last day (0.74929 ± 0.08208 mA). These electric current values are due to the good formation of the electrogenic biofilm, and according to Mbugua et al. (2020), this is highly dependent on the carbon sources present in the substrates [36]. Likewise, it has been shown that high levels of carbohydrates serve as the main sources of carbon for microbial activity, causing the production of electrons that flow from the anodic to the cathodic electrode that generate electrical current [37,38]. The electrical current peaks coincide with those generated by other authors using citrus in MFCs with a single chamber, where they explain that the decrease in current values is due to the fact that the fresh substrate is exhausted and a sediment begins to form in the upper part of the MFCs, thus generating a decrease in the compounds [39].

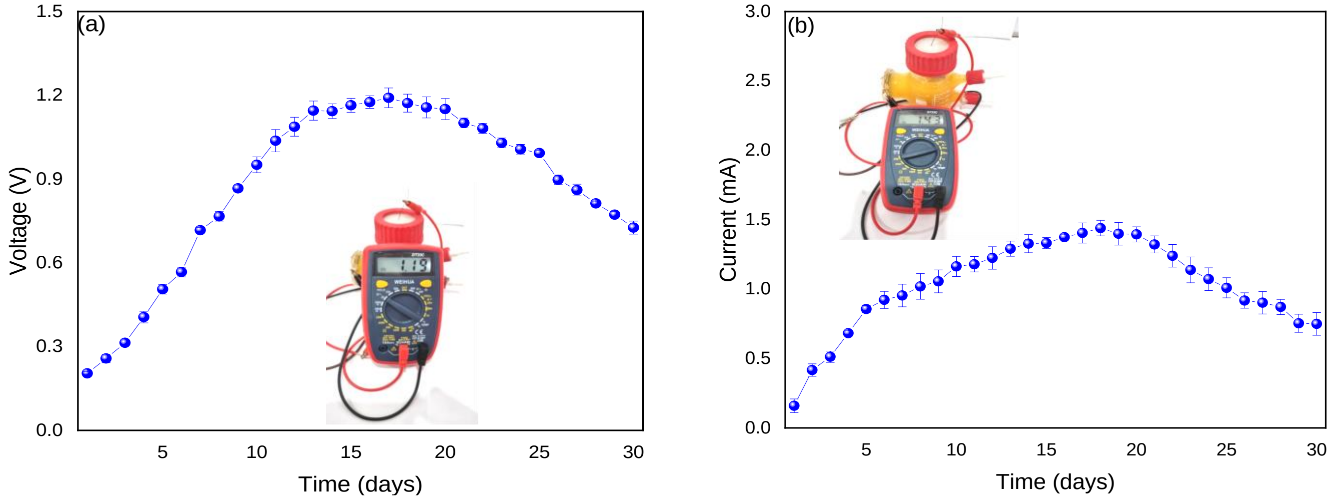

Figure 2. Values of (**a**) voltage and (**b**) electrical current of microbial fuel cells.

Figure 3a shows the monitored pH values during the 30 days; it was observed that the values remain in the slightly acidic region, with the optimum operating pH of 4.78 ± 0.46 on the seventeenth day. The pH values increased during monitoring due to the fermentation of the substrate used, which according to Igboamalu et al. (2019), all MFCs have a saturation potential that depend directly on the pH because the consortium of microorganisms that can exist within a substrate needs a suitable pH for growth [40]. Although investigations have been reported that have managed to generate electrical values with neutral or basic pH, the values obtained by our investigation are among the highest shown (without adding chemical compounds to the substrate). For example, Prasidha W. (2020) works their MFCs at a pH of 7.1 using food waste leachate as a substrate, managing to generate voltage peaks of 410 mV [41]. Likewise, Figure 3b shows the values of electrical conductivity, where the values increase from the first day (70.46 ± 1.73 mS/cm) to day 18 (140.07 ± 3.51 mS/cm) and then decrease slowly until the last day (45.98 ± 4.51 mS/cm). The increase in electrical conductivity values is due to the low electrical resistance of the substrate used, while these values begin to decline due to the sedimentation of organic compounds present in the waste used [42,43]. Substrate masses have also been reported to have a dependence on electrical conductivity According to Kalagbor et al. (2020), this relationship is directly proportional because in their research, the masses increased from 1 to 12 Kg and their electrical conductivity values increased from 787.6 ± 475.89 to 1282.9 ± 492.94 mS/cm. Said increases were also represented by the values of voltage and electric current [44]. In Figure 3c, the ° Brix values monitored for 30 days are shown, and it can be observed that they gradually declined from the third day (14° Brix) until the last day (0° Brix). Tangerine is a fruit that is composed of almost 76% moisture and 24% soluble solids (such as sugar-fructose and glucose) [45], which are precisely the rich sources of carbons that microorganisms use for their growth [46]; this would be one of the important characteristics of the decrease in the Brix degree values in the bioelectricity generation process [47].

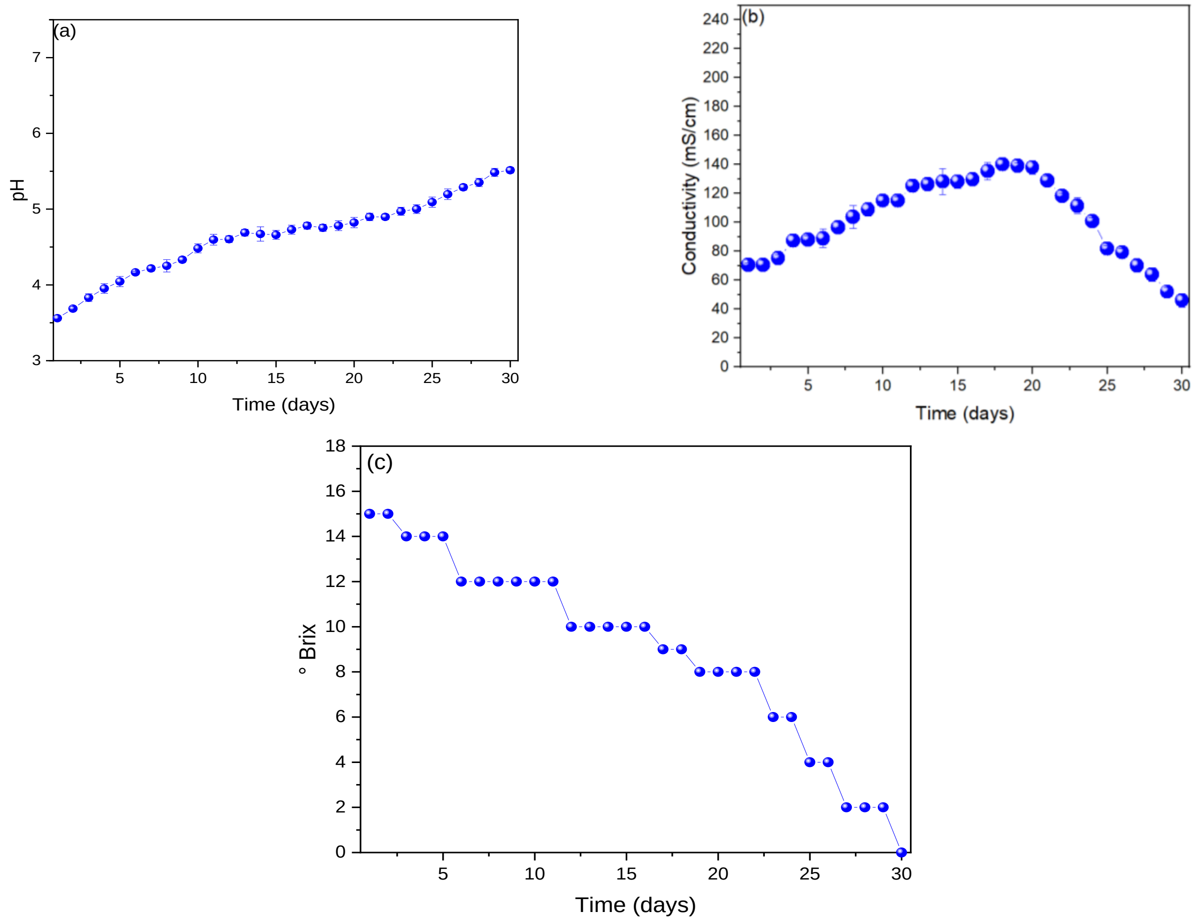

Figure 3. Monitoring of the values of (**a**) pH, (**b**) conductivity and (**c**) Brix degrees of the microbial fuel cells.

Figure 4a shows the internal resistance ($R_{int.}$) of the microbial fuel cells, for which Ohm's Law (V = IR) was used. The voltage values were placed in "Y" and those of current in "X", and in this way the slope found using the linear fit represents the internal resistance of the MFCs. The calculated $R_{int.}$ value was 65.378 ± 1.967 Ω; this value was calculated at the maximum peak of voltage and electric current generation (seventeenth day). Lower internal resistance values than those shown in this investigation have been reported, but with lower voltage and current values as well, although the theory would indicate that for lower resistance the electrical current values should be higher [48,49]. For example, Van Der Velden et al. (2022) calculated an internal resistance of 15.99 Ω in their MFCs using sediments from Saldanha Bay, which operated at pH 9 and used carbon electrodes [50]. In this same sense, Torlaema et al. (2022) found an internal resistance of 38.87 Ω in their single-chamber MFCs, managing to generate voltage and current peaks of 168 mV and 0.168 mA using rice waste and graphite electrodes as a substrate [51]. Figure 4b shows the power density (PD) values in the current density (CD) of the MFCs, managing to show PDmax peaks of 475.32 ± 24.56 mW/cm^2 in CD of 5.539 A/cm^2 with a peak voltage of 1024.12 ± 25.16 mV. Other authors reported better values than those we show; for example, Rokhim et al. (2022) managed to generate PD peaks of approximately 90 mW/cm^2 in their single-chamber MFCs using banana debris as a substrate. According to the authors, the increase in organic matter (substrate) in the anodic chamber increases the PD values [52]. Likewise, Yaqoob et al. (2022) managed to generate PD peaks of 0.22, 0.30 and 0.71 mW/cm^2 with internal resistances of 380, 450 and 560 Ω, managing to demonstrate that the values of the internal resistances of the MFCs imparted on the PD values, and these values of the internal resistances are being influenced by the sizes of the electrodes used [53].

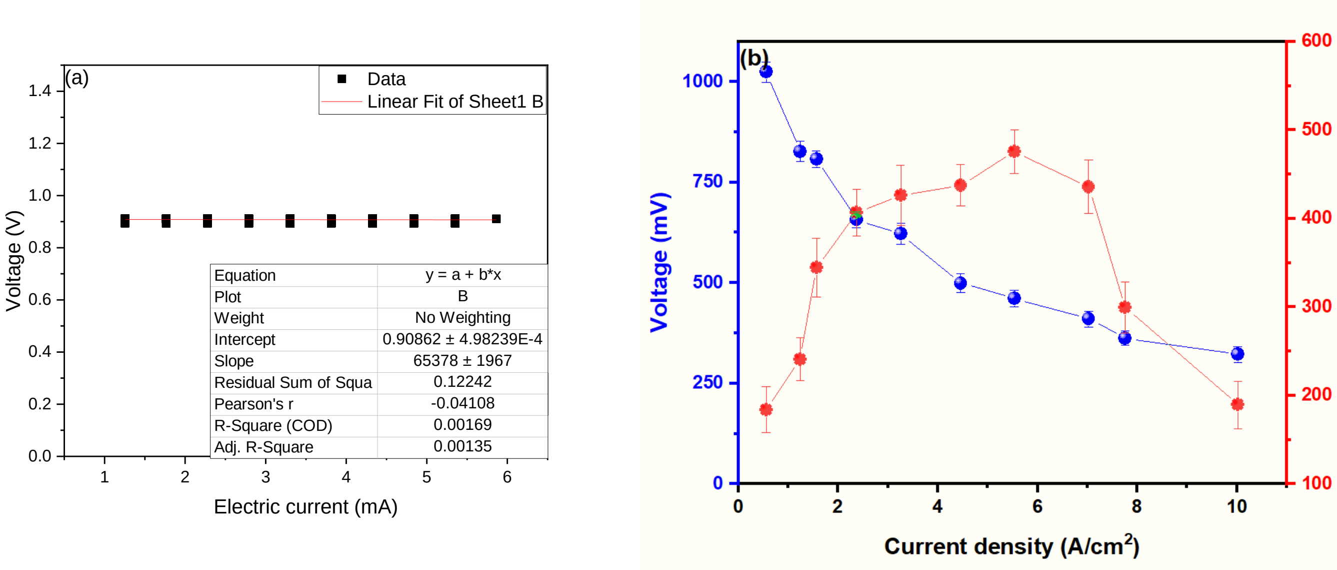

Figure 4. Values of (**a**) internal resistance and (**b**) power density as a function of current density.

Table 1 shows two species of microorganisms isolated and identified from orange residues. The identification was possible through the molecular biology technique which, according to the characterization in the Blast program, found the bacteria and yeast *S. fonticola* and *R. mucilaginosa* with an identity percentage of 99.57 and 99.50%, respectively. The first species belongs to a bacterium within the phylum Proteobacteria, while the second microorganism belongs to a yeast within the phylum Basidiomycota. The enterobacterium *S. fonticola* is ubiquitous in nature, that is, it can be found surviving in various habitats, and some species cause food spoilage [54,55]. Due to this, it is possible to find them in orange residues in the same way the yeast *R. mucilaginosa* is ubiquitous and can be found in organic residues from which it can generate pigments [56,57].

Table 1. Species identified from the anode of the MFCs with pitahaya residues.

Waste	Coding	Identified Species	Type of Microorganism	pb	% of Identity	Access Number
tangerine	5L	*Serratia fonticola*	Bacterium	1387	99.57	NR_025339.1
	9L	*Rhodotorula mucilaginosa*	Yeast	611	99.50	NR_073296.1

The isolation of different types of microorganisms (Bacteria, Archaea and Eukaryas) is possible from substrates contained in MFCs as indicated by Greenman et al. (2021) [50]. On the other hand, it is possible to isolate mostly proteobacteria which are electrogenic within MFCs [58,59]. Similarly, it is known that yeasts are more active within an MFC than bacteria with respect to electron transfer [60]. In this sense, both *S. fonticola and R. mucilaginosa* may be associated with the generation of bioelectricity through the oxidation of organic residues (orange residue) [61]. In Thulasinathan et al. (2021), it was shown that Serratia marcescens AATB1 can form biofilms on the electrode surface of an MFC [62]. In Ali et al. (2020), Serratia marcescens was identified in a microbial community of an MFC that had microalgae biomass as a substrate [63]. Regarding the yeast *R. mucilaginosa*, it is known that it produces carotenoids from food residues [64], which may be associated with the transfer of electrons to the electrode. This can be supported by Shrestha et al. (2016), who mention that some species of compounds with redox activity, such as carotenoids, may be involved in the generation of electricity [65]. Figure 5 shows the schematization of the electric current generation process through microbial fuel cells, which were connected in series, managing to generate 3.15V—enough to light an LED light (red).

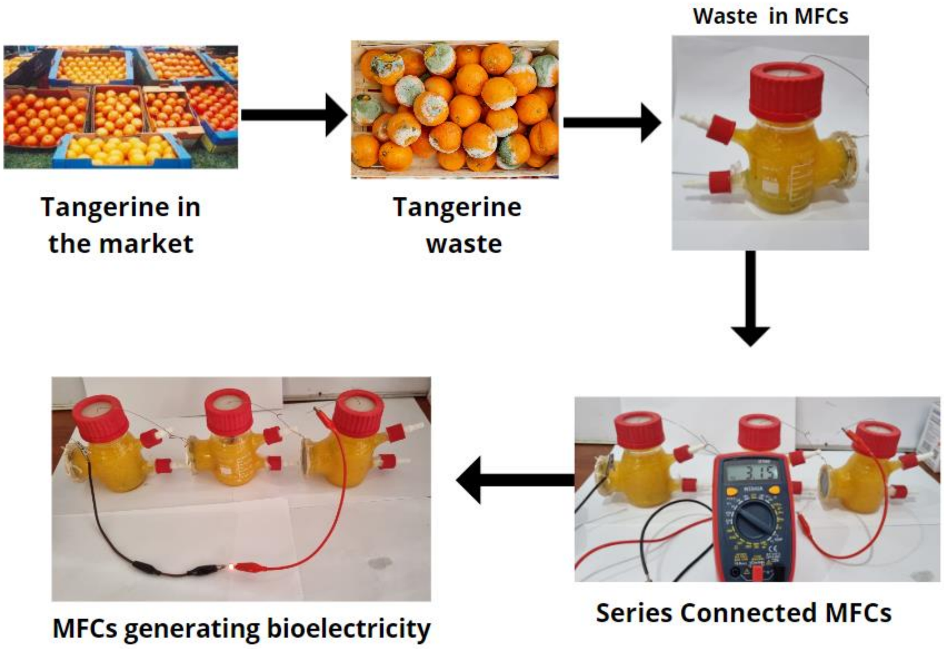

Figure 5. Diagram of the bioelectricity generation process.

Table 2 shows the electrical parameters obtained in other investigations with similar debris found in the literature, where it can be seen that the voltage values (1.191 ± 0.035 V) found in this investigation exceed those shown in the literature. Mishra (2023) and Rincón et al. (2022), where they worked at an acidic pH but with carbon or graphite electrodes, managing to generate voltage peaks and a power density lower than those shown in this research [26,66].Although the values found by Flores et al. (2020) and Rojas et al. (2022) are very close to those reported in this research, this may be due to the electrodes used (metallic in these cases) that helped the flow of electrons within the MFC [67,68]. On the other hand, Asefi et al. (2019) generated 0.600 ± 0.025 V using food remains as a substrate, which have a high amount of carbon in their composition but work at a slightly alkaline pH, and the electrodes used were graphite [69]. This is repeated in the investigations carried out by Verma.

Table 2. Electrical parameter values obtained in MFCs published in the literature.

Substrate Type	MFC Type	Maximum Voltage (V)	Power Density (PD) (W/m^2)	Current Density (CD) (mA/cm^2)	Reference
Sweet Lemon Peels	dual-chamber	0.792 ± 0.0153	204.80 ± 1.28	640.0 ± 2.0	[66]
Banana Waste	Singler chamber	0.286	41.3	286.7	[21]
Lime, orange and tangerine waste	Singler chamber	0.99 ± 0.089	0.0628	0.049	[67]
Blackberry, dragon fruit and noni	Singler chamber	0.97 ± 0.12	0.0719 ± 0.0012	0.051	[68]
Food waste	Dual chamber	0.600 ± 0.025	0.345	830	[69]

4. Conclusions

Bioelectricity was successfully generated using pilot-scale microbial fuel cells using tangerine waste and low-cost electrodes (copper and zinc) as fuel, managing to generate voltage and electric current peaks of 1.191 ± 0.035 V and 1.43973 ± 0.05568 mA on days 17 and 18, respectively. These values were obtained while operating in slightly acidic pH regions, with an optimum operating pH of 4.78 ± 0.46 and with a peak value of electrical conductivity of the substrate of 140.07 ± 3.51 mS/cm, while the values of Brix degrees decreased slowly until the last day (zero ° Brix). Likewise, an internal resistance of 65.378 ± 1.967 Ω was found in microbial fuel cells, with a maximum power density of 475.32 ± 24.56 mW/cm^2 at a current density of 5.539 A/cm^2 and a peak voltage of 1024.12 ± 25.16mV. From the biofilm formed on the anode electrode, it was possible to identify the bacteria and yeast *S. fonticola* and *R. mucilaginosa* with 99.57 and 99.50% identity. Finally, the microbial fuel cells were connected in series, generating 3.15 V and managing to turn on an LED light (red). The potential of citrus fruits such as tangerines in the generation of electrical energy at the laboratory level has been demonstrated, as well as showing a process that allows the mitigation of the impact of solid waste on the environment.

One of the main limitations on this research is the cost of the microbial fuel cells manufacturing materials. Although the selected electrodes are relatively low cost for this research, it is still necessary to design other models and use other materials. Therefore, taking them to a larger scale is not feasible, so it is still premature to carry out a general economic analysis at this stage of the investigation.

For future work, it is recommended to add glucose or sucrose, because there are investigations where they have managed to increase the electrical parameters up to a certain saturation point. It is also recommended to work at the optimum pH found in this investigation and with electrodes covered with non-toxic material for electricity-generating microorganisms. Increasing the volume of microbial fuel cells would help increase the potential for electric power generation.

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