



## The effect of nutrient broth media on PEDOT:PSS gated OECTs for whole-cell bacteria detection

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### ABSTRACT

The organic electrochemical transistor (OECT) is a device of much interest in biological sensing applications, including the detection of various cell types and electrolytes. In order to study cell behavior these devices are often operated in a cell culture media. While OECT operation in various salt solutions is well documented the effects of nutrient media are rarely addressed. We have investigated the effect of the complex nutrient broths commonly used in cell culturing on the OECT operation. Here we report on the effects of using lysogeny broth (LB) and yeast/tryptone (YT) nutrient broths as operating media on PEDOT:PSS based planar OECTs. OECT devices were characterized in both nutrient broths and equivalent salt solutions to determine the effects that proteins and yeast extracts have on current on/off ratio, transconductance, and gate voltage at peak transconductance. We have found that the OECT peak transconductance ( $\sim 2.4$  mS) was not affected by operation in broth media. However, a shift of  $-250$  mV in effective gate voltage was observed. This resulted in a reduction in the gate voltage for peak transconductance from  $\sim 0.7$  V for a pure electrolyte solution to  $0.3$  V for operation in both broth solutions. A doubling in the ratio of operating current (at peak transconductance) to initial current from  $\sim 0.2$ – $0.3$  to  $\sim 0.5$ – $0.6$  was observed for both broth media. To demonstrate application as a biosensor in nutrient media, the OECT was used to characterize and detect the bacterium *Pseudomonas fluorescens*, achieving a limit of detection of  $\sim 880$  CFU/mL while operating in LB broth.

### 1. Introduction

One of the commonly used devices in biological characterization and detection is the micro-electrode array (MEA) (Arunasri and Mohan 2019). These sensors are popular due to their ease of use, simple fabrication, ability to integrate into flow systems, and adjustable transduction materials (Zhang et al., 2013). Unfortunately, MEA signal strengths (either impedance, current, or voltage) typically require sensitive driving electronics with external signal amplification. To help address this complication, the use of transistors as biosensors combines most of the benefits of the MEA with the addition of built-in signal amplification (Piro et al., 2018). Electrolyte-gated transistors, such as the organic electrochemical transistor (OECT), operate in an electrolyte medium and can be used to interface with biological signals in the same way as MEAs.

Over the past decade, significant research has been reported using OECT in biosensing (Ahmed et al., 2014; Lin et al., 2010b; Marks et al., 2022; Piro et al., 2018). OECTs have been used for whole cell characterization (Hempel et al., 2017) and detection (He et al., 2012), redox

reactions (Galliani et al., 2020; Guo et al., 2019), ion-sensing (Sessolo et al., 2014), enzymatic sensing (Yang et al., 2010), and other biological applications (Strakosas et al., 2015). These devices are popular due to their unique structure providing a biocompatible interface, high transconductance, and ability to convert ionic signals into electronic signals (Donahue et al., 2018). These traits come from the use of an ion-permeable semiconducting channel, typically a conductive polymer such as PEDOT:PSS.

In an OECT applied gate voltage drives electrolyte ions into the channel and modulating the source-drain channel current ( $I_{DS}$ ). In the case of PEDOT:PSS the ions de-dope the channel, and the OECT behaves as a depletion-mode transistor (Rivnay et al., 2018). The source-drain current for these devices is then given by eq. (1) for operation in the linear regime and eq. (2) in saturation regime (Bernards et al., 2008; Bernards and Malliaras 2007). Here,  $q$  is electron charge,  $\mu$  is the hole mobility,  $p_0$  is the hole concentration,  $t$  is the thickness of the semiconductor layer,  $w$  is the channel width,  $L$  is the channel length,  $V_p$  is the pinch-off voltage  $V_G^{eff}$  is the effective gate voltage, and  $V_{DS}$  is the drain-source voltage.

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$$I_{DS} = \frac{q\mu p_0 t w}{L V_p} \left( V_p - V_G^{eff} + \frac{V_{DS}}{2} \right) V_{DS} \text{ for } V_{DS} \ll |V_p - V_G^{eff}| \quad (1)$$

$$I_{DS} = \frac{q t w \mu p_0}{L V_p} (V_G^{eff} - V_p)^2 \text{ for } V_{DS} \geq |V_p - V_G^{eff}| \quad (2)$$

The usage of an effective gate voltage is derived from the OECT's operation in an electrolyte media. This causes formation of electric double layers (EDL) at the gate-solution interface and the solution-channel interface. Introduction of an offset voltage ( $V_{offset}$ ) at either interface can result in a shift in source-drain current, which is interpreted as a change in the effective gate voltage (eq. (3)).

$$V_G^{eff} = V_G + V_{offset} \quad (3)$$

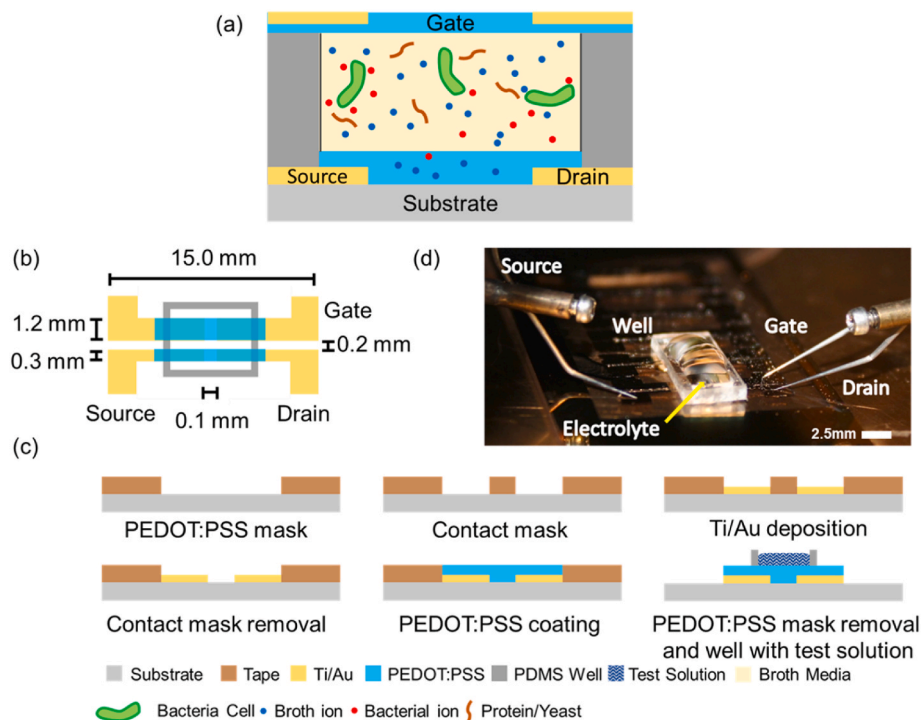
These offset voltages can be created by changes in pH (Scheiblin et al., 2017), ion-concentration (Lin et al., 2010a), redox reactions (Bernards et al., 2008), or contact with a charged particle such as the membrane of a cell (Demuru et al., 2019).

For any given approach, the OECT design and operation are optimized for its chosen sensing mechanism and application (Ramuz et al., 2015; Yaghmazadeh et al., 2011). One of the variables in OECT operation is the choice of operating media. OECTs have been operated in a wide range of media including pure electrolyte solutions, phosphate buffered solutions (PBS), nutrient broths (Demuru et al., 2019; Guo et al., 2019; Lingstedt et al., 2019; Yeung et al., 2019), blood and plasma (Preziosi et al., 2022), and sea water (Liao et al., 2014) (Supplemental Table S1). The operating medium can even be in the form of a gel (Khodagholy et al., 2012). With the wide range of operating media available to the OECT the performance of the transistor can vary greatly without changing the structure of the device.

The effects of ion concentrations on OECT operation have previously been well studied (Lin et al., 2010a). However, the effects of complex media on OECT operation are not typically defined or discussed. The understanding of OECT operation for a given testing media can allow for better optimization of transistor geometry (Yeung et al., 2019) and testing parameters. Despite its usage in both cell culturing and OECT

sensing, studies into the effects of complex nutrient broths on OECT performance have not previously been reported.

In this work, we investigated the effects of two well established bacteria broths on OECT performance. OECT operation was characterized in  $2 \times$  yeast extract-tryptone (YT) broth and lysogeny broth - Miller (LB) nutrient broths, both typically used for the culture of bacteria (Fig. 1a). By comparing against saline solutions with equivalent ion concentrations, we can identify the effects of the components unique to the broth media. Determining the effects of proteins and yeast extracts on these parameters allow us to adjust the device design for optimum sensitivity in the operating media. The gate voltage for peak transconductance is an operating parameter that determines the optimal biasing condition for the sensor. By identifying how the OECT peak gate voltage in the culture broths differs from that of pure electrolyte solutions, one can tune the measurement parameters for biological detection and characterization. Broth media were tested as liquids, as well as agar gel electrolytes, both common forms in bacteria culturing. Devices were characterized through current-voltage (I-V) curve trace measurements, as well as time response measurements. The optimized OECT is then utilized to detect and characterize the presence of bacteria in nutrient broth. This is accomplished by measuring the change in source-drain current induced on the OECT by the membrane potential of bacteria. In this approach, a capture mechanism is not used to attach the bacteria to the OECT. Instead, we rely on detecting the bacteria through their natural processes, such as ion generation and spontaneous attachment to the channel and/or gate of the device as they begin to precipitate in the well (Flemming et al., 2016). The bacterial membrane potential is reported (Demuru et al., 2019; He et al., 2012) to be the main contributor in OECT detection of bacteria. This approach allows for "universal" (non-specific) detection of the bacteria and will work in principle with any bacteria type that has a membrane potential and that will adhere to PEDOT:PSS (Gomez-Carretero et al., 2017; Mehes et al., 2020). The challenge for this approach is that the baseline signal is dependent on determining OECT operation in the test media. Detection of bacterial contamination is achieved upon observing deviation from the baseline (sterile case) signal. The overall measured signal in the uncaptured



**Fig. 1.** (a) General approach for utilizing OECT for uncaptured bacteria detection and culture media characterization; (b) planar OECT layout; (c) fabrication steps using a two-stage tape mask liftoff technique; (d) photo of OECT device under test.

approach is a combination of contributions of multiple signal sources additive or subtractive effect, such as bacteria attachment to the gate and/or channel. In our approach the bacteria interact spontaneously with the electrodes, therefore the sensing characteristics will depend on the state of the bacteria. The state will depend on several factors, including temperature, media chemistry, type of bacteria, and metabolic state. Another factor that could affect the of sensing properties is the presence of additional particles in the media, particularly charged particles. The bacterium *Pseudomonas fluorescens* was chosen for investigating OECT response to uncaptured bacteria due to its safe handling and relation to the antibiotic resistant strains of *Pseudomonas aeruginosa*. All bacterial detection was performed in LB broth culture media at various concentrations, without the addition of other diluents. To our knowledge, the effects of various nutrient broths on OECT operating have not been previously reported. This is a critical step towards characterizing and understanding OECT sensing of dispersed bacteria.

## 2. Design and fabrication

### 2.1. Design

To test the effects of bacterial broth media on OECT operating parameters we chose a simple planar design. The layout of the structure is similar to that of previously reported all-PEDOT:PSS OECTs (Ramuz et al., 2015), wherein both the channel and gate consist of PEDOT:PSS. Use of PEDOT:PSS for both the gate and channel allowed for simplified fabrication as well as a biocompatible interface on both electrodes. An Au electrode beneath the PEDOT:PSS was utilized to create a defined channel region and source/drain contacts. The key advantage of this planar structure is that it is simple to fabricate and does not need an additional gate electrode suspended within the electrolyte solution. Dimensions for the transistor are shown in Fig. 1b.

### 2.2. Fabrication

OECTs were fabricated in a 6-transistor array on a solvent-cleaned 25 mm × 75 mm glass microscope slide. Device patterning was performed using a two-stage tape mask liftoff technique (Fig. 1c). Both tape masks (PEDOT:PSS mask and contact mask) were applied onto the microscope slide, with the contact mask resting over the PEDOT:PSS mask. The masked sample is then cleaned further with O<sub>2</sub> plasma for 2 min. Ti (adhesive layer) and Au (conducting layer) were then sequentially sputtered to form ~50 nm thick source-drain and gate electrodes. The contact mask is then removed from the sample, establishing the channel gap between source and drain electrodes. The PEDOT:PSS (1.3 wt%, Sigma Aldrich) layer (~100 nm) was then applied via a doctor blade. Next, the devices were annealed at 120 °C for 30 min following procedures used in literature (Sessolo et al., 2013). The samples were then cooled to room temperature before removal of the PEDOT:PSS mask. Later devices that were used for characterization of various bacterial concentrations (CFU/mL) were post-treated with a DMSO solution to increase PEDOT:PSS conductivity and, hence, device transconductance from ~2 to ~20 mS. The post-treatment followed the procedure from Lue et al. (Luo et al., 2013). Devices were covered in DMSO for 30 min then dried with N<sub>2</sub> gas and finally baked at 130 °C for 30 min. Finally, a laser cut PDMS well was placed over two adjacent devices to contain the electrolyte for testing (Fig. 1d).

## 3. Methods

OECT operating parameters were measured with LB and YT culture media and for comparison with pure NaCl solutions. NaCl solutions with molar concentrations of 1.0, 0.17, 0.1, 0.085, and 0.01 M were used in testing. Both NaCl and nutrient solutions are prepared with a pH of 7. LB and YT broths were of particular interest as they would allow the characterization of the OECT response to bacteria in their native growth

media. These broths were also chosen due to their relatively simple ingredient list. Recipes for both YT and LB broth contain only NaCl, tryptone, and yeast extracts in different concentrations. The salt present in both media enables OECT operation. Effects of broth components (proteins and yeast extracts) in the media are then monitored in the electrical characterization of the OECT. For a direct comparison, the broth media were compared against salt solutions containing the same concentration of NaCl as the broth (0.17 M for LB, 0.085 M for YT). The conductivity of nutrient media (16.9 mS/cm for LB broth and salt solutions (16.3 mS/cm) were comparable. The full recipes for both broths can be found in Supplemental Table S2.

### 3.1. OECT characterization methods

Characterization of OECT operation with different operating media was performed using I–V curve traces (Fig. 2a). A test solution of 40 μL was dispensed into the PDMS well, covering two OECT sensors, and allowed to rest for 15 min prior to testing. For agar-based devices, 40 μL of heated broth agar (180 °C) was dispensed into the PDMS well. The agar was then covered with a glass cover slide and allowed to cool for 1 h before testing. After media dispensing curve traces were performed by sweeping the drain - source voltage ( $V_{DS}$ ) at various values of the gate voltage ( $V_G$ ) and measuring the drain - source current ( $I_{DS}$ ). The gate voltage was applied using an adjustable DC power supply from 0.0 V to 1.0 V in 0.1 V increments. Drain-source voltage application and current measurements were performed using a HP 4140B Picoamp meter/DC voltage supply. Drain - source voltage from 0.0 V to -1.0 V was incremented in 0.05 V increments every 4 s.

Three operating parameters were extracted from the I–V characteristics: the ratio of operating current to initial (zero bias) current ( $I_{OP}/I_0$ ), peak transconductance ( $g_{m,peak}$ ), and the gate voltage required for peak transconductance ( $V_{G,peak}$ ) (Fig. 2b). The transconductance plays the key role in determining device sensitivity.

### 3.2. Bacterial culture preparation

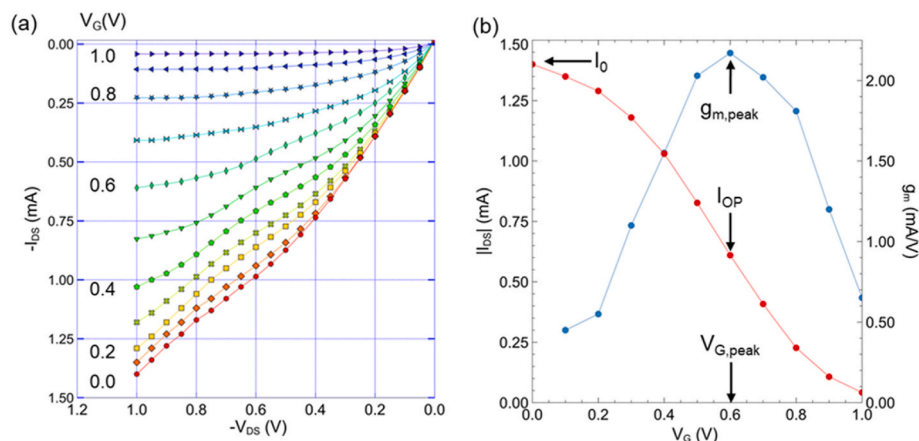
Bacterial solutions were prepared using a fixed growth procedure. First, *Pseudomonas fluorescens* bacteria from a plate culture were inoculated into 5 mL of LB growth media. The liquid culture was then incubated at 32 °C for 48 h, after which it is used for two different serial dilutions, one in PBS and the other in LB broth. Dilution in PBS is used to establish the CFU/mL of the grown culture (using a standard cell counting technique), while dilution in LB broth is used to create desired CFU/mL concentrations for OECT testing. LB broth was chosen as the diluent for OECT testing in order to reduce potential effects of media dilution on OECT characteristics.

## 4. Results

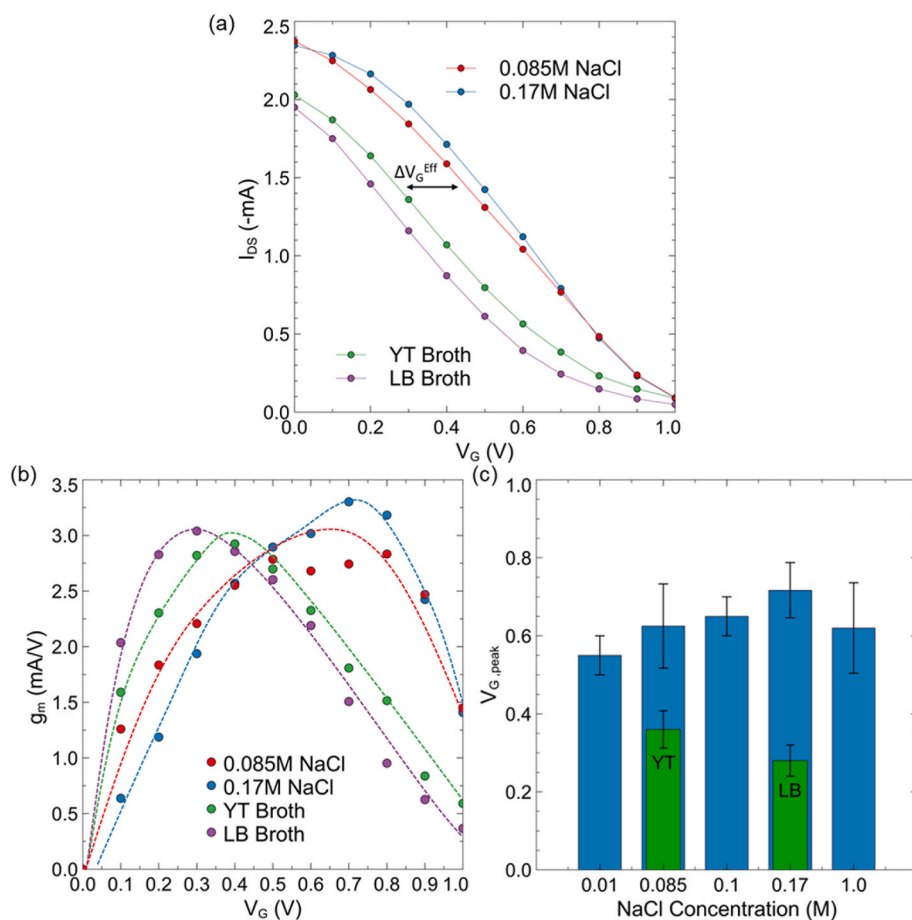
### 4.1. OECT operation in nutrient media

Differences between OECT operation in broth and their equivalent salt solutions can be seen in their respective transfer I–V ( $I_{DS}$  vs  $V_G$ ) characteristics (Fig. 3a). These results show that OECTs operated in broth media have a lower source drain current than their equivalent salt counterparts. The similar transconductance values (Supplemental Table S3) between broth and salt solution indicate that the decrease in current is due to the increased effective gate voltage. The shift in the broth media compared to their equivalent salt solutions indicate a ~0.25 V increase in effective gate voltage when operated in broth media.

This shift in effective gate voltage can be seen more clearly when transconductance is plotted against gate voltage (Fig. 3b). This shows a gate voltage shift for peak transconductance from 0.7 V for 0.17 M NaCl to 0.3 V for LB broth, and from 0.8 V in 0.085 M NaCl to 0.4 V in YT broth. In both cases the use of bacteria broth shifted the peak



**Fig. 2.** Typical OECT electrical I-V characteristics in NaCl (0.17M) solution: (a)  $I_{DS}$  vs  $V_{DS}$  (0.0 to  $-1.0$  V) curve traces at  $V_G$  values from 0.0 to 1.0 V in 0.1 V increments; (b) transconductance and drain-source current as a function of gate voltage at  $V_{DS} = -1.0$  V;  $V_{G,peak}$  is the value of gate voltage at which the maximum transconductance ( $g_{m,peak}$ ) is obtained.



**Fig. 3.** (a) OECT drain - source current at  $V_{DS} = -1.0$  V for operation in LB and YT nutrient broths and their equivalent pure NaCl solutions. (b) OECT transconductance as a function of gate voltage for devices operated in LB and YT nutrient broth media compared against equivalent salt solutions. (c) Gate voltage required to achieve peak transconductances ( $V_{G,peak}$ ) for pure salt solutions (blue bars) and nutrient broths (green bars). (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

transconductance voltage by  $\sim 0.4$  V, higher than the 0.25 V observed in Fig. 3a. This change indicates that operation in the broth media does not simply shift the effective gate voltage, but also changes the slope of the transconductance.

The large change in effective gate voltage is most likely the result of a redox reaction between PEDOT:PSS and tryptone and yeast extract present in the broth (Ullmann and Bombarda 2013), but absent in the NaCl solutions. A similar shift in  $V_G^{eff}$  is reported for devices that used redox reactions at the gate sensing electrode for glucose and bacteria

(Butina et al., 2019; Shim et al., 2009). Redox reactions at the gate electrode result in a decrease in the EDL, resulting in an increase in  $V_G^{eff}$  on the channel.

The differences in  $V_{G,peak}$  for operation in pure NaCl of various concentrations and in broth solutions are evident in Fig. 3c. These results indicate a shift of roughly  $-0.3$  V between operation in broth media and in NaCl solutions, similar to that seen in Fig. 3b. Very low  $p$  values have been calculated for broths and equivalent NaCl solutions: 0.014 (between 0.085 M NaCl and YT broth),  $\sim 0.0005$  (between 0.17 M NaCl

and LB broth). Differences between the salt solutions are minimal and hidden by the standard deviation between devices. This is consistent with the 57.6 mV/dec shift for  $\text{Na}^+$  concentration reported in literature (Lin et al., 2010a). This indicates that the difference between the two types of media is due to the presence of biomolecules in the broths and not the salt content of the solutions.

The shift in  $V_G^{\text{eff}}$  due to the broth components acts as a background signal that should be accounted for when designing OECT parameters. Differences in magnitude of effective gate voltage shift seen in the LB vs YT media could be due to the differences in concentration of the yeasts and peptides in either formula. The major advantage of the increased effective gate voltage for bacteria sensing is the ability to operate the OECT at a lower applied gate voltage (Rivnay et al., 2013). This is useful to reduce the effects of electrode voltage on cell behavior, such as bio-film formation (Gomez-Carretero et al., 2017) and surface attachment (Arunasri and Mohan 2019).

#### 4.2. Agar OECT switching response

Additionally, OECT switching time measurements were performed using the broth in solution and as an agar-gel. These measurements were performed by pulsing the gate voltage at a fixed drain-source voltage ( $-1.0$  V). The OECT was switched between on and off states by toggling the gate voltage between 0.0 V (on state) and 0.3 V (off state), with each state lasting 5 s. The source-drain current was measured at 50 ms intervals to record the rising and falling times corresponding to the on-and-off switching processes. The switching time was defined as the time required, after introducing or removing gate voltage, for the source-drain current to reach within 5% of its steady state value for both on-off and off-on switching. The recorded switching time for each case is an average over 6 on-off cycles for each testing medium.

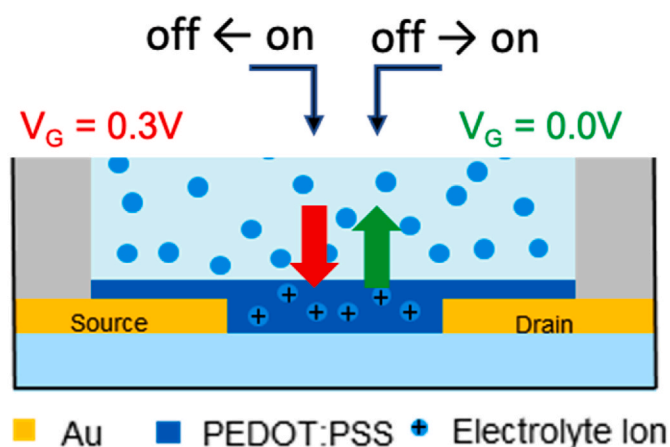
Switching times for solution and agar broths are shown in Table 1. The switching-off times ( $t_{\text{off}}$ ) of the YT agar devices were found to be much longer than that of its solution counterpart, while the switching-off time of the LB agar was only marginally longer than that of the solution. Interestingly, the switching-on time ( $t_{\text{on}}$ ) did not show this trend. Switching-on speeds for agar and solution devices are within 1–2 standard deviations of each other, with agar devices switching slightly faster. The switching-on times for both agar and solution devices were notably longer than the switching-off times.

This difference in switching times can be explained based on the estimated ion mobility in each media. For the switching off process, the ions move predominantly from the electrolyte media into the polymer channel material (Fig. 4 red arrow). Ions are expected to have a significantly lower mobility in the agar compared to liquid media. This correlates with the observed increase in switching-off times seen in the agar devices. In contrast, during the switching-on process ions move from the PEDOT:PSS layer back into the electrolyte (Fig. 4 green arrow). In this case the media that the ions are traveling through PEDOT:PSS is identical in both agar and solution cases. This correlates well with the observed results of switching-on being similar between agar and solution media. The longer switching-on versus switching-off times also indicates that ion mobility in the PEDOT:PSS is lower than that in either the agar or solution media. The slightly lower switching-on times for

**Table 1**

OECT switching-on and -off times for LB and YT solutions (broth) and agar-gel operating media. Two devices were measured at least 3 times for each medium type. Time base measurements had a measurement interval of 50 ms due to equipment limitations.

Medium	$t_{\text{off}}$ avg (ms)	$t_{\text{off}}$ std dev (ms)	$t_{\text{on}}$ avg (ms)	$t_{\text{on}}$ std dev (ms)
YT Broth	$\leq 50$	$\leq 50$	310	23.5
LB Broth	75	43.3	325	70.7
YT Agar	200	37.7	285	51.5
LB Agar	100	46.9	258	18.6



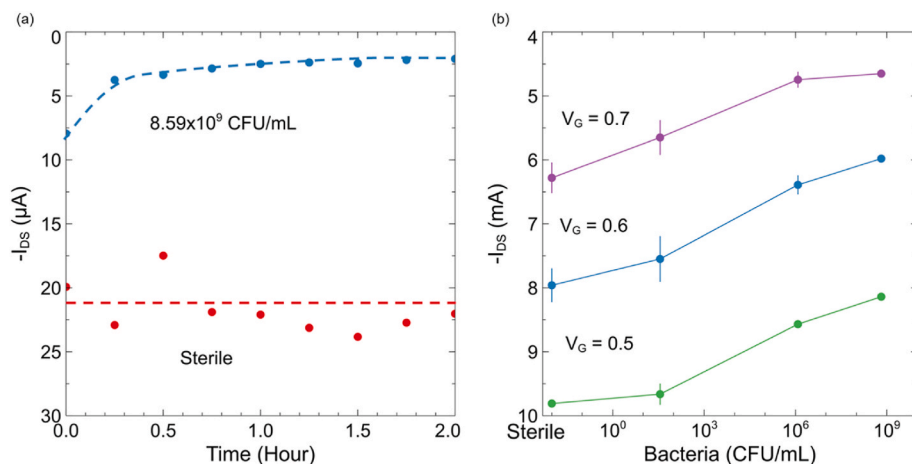
**Fig. 4.** Movement of ions into and out of OECT polymer channel under applied bias: switching states from on to off (applying voltage, red arrow) and off to on (removing voltage, green arrow). (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

agar devices compared to solution media can potentially be a result of diffusion assisted transport out of the PEDOT:PSS channel that is not present in the solution media. During switching-off the agar media near the channel becomes depleted of ions. This depleted region can then assist in switching-on by providing a diffusion gradient to extract ions from the PEDOT:PSS. This is not seen in the solution case due to the higher ion mobility in solution filling the depleted region before the OECT switches back on. Confirmation of these hypotheses can be obtained with additional measurements and studies on ion mobilities in comparable agar and solution medias.

#### 4.3. OECT uncaptured bacterial response

The OECT response to bacteria in LB broth media was measured as a function of time to demonstrate the ability to measure potential bacterial growth and surface adhesion. To achieve this the source-drain current was continuously monitored over a 2-h period in *Pseudomonas fluorescens* solutions containing  $8.59 \times 10^9$  CFU/mL. Measurements in sterile LB were carried out in parallel. During the test the gate voltage was held at the measured peak transconductance point for LB of 0.3V. To prevent sample evaporation, the PDMS wells were covered with a glass microscope slide after solution dispensing. Fig. 5a shows a clear distinction between the sterile and bacteria containing broth media. The bacteria-containing broth experiences much lower currents than the sterile broth equivalent, indicating an increase in effective gate voltage, as expected in the operation of a depletion-mode transistor (Bernards and Malliaras 2007). The bacteria-containing sample also requires a longer time to reach equilibrium, indicating that the OECT is responding to continued attachment of bacteria to the surface of the device. Additionally, the current for the bacteria containing sample continues to slowly increase after the initial half hour period, potentially due to bacteria growth on the device and/or in the media.

Operation of the OECT as a bacterial detection sensor in LB broth was also evaluated at several different concentrations of bacteria. This was achieved by measuring DMSO post-treated OECT  $I_{\text{DS}}$  at several gate voltages ( $V_G = 0.5$  V, 0.6 V, 0.7 V) for different concentrations of bacteria. Bacteria samples were prepared from a culture in LB broth ( $\sim 5.0 \times 10^9$  CFU/mL) and then serially diluted in sterile LB broth to the desired concentrations for testing:  $3.6 \times 10^7$ ,  $1.2 \times 10^6$ , and  $6.6 \times 10^8$  CFU/mL. The DMSO post-treatment significantly increased the on-current compared to previous OECTs (used in Fig. 3), resulting in improved transconductance. The results (Fig. 5b) show a decrease in drain source current with increasing bacteria concentrations starting in the  $10^2$ – $10^3$  CFU/mL range, consistent with results seen in the time



**Fig. 5.** OECT characteristics: (a) time response to sterile LB broth as operating media (red) vs cultured LB broth containing  $8.59 \times 10^9$  CFU/mL of *Pseudomonas fluorescens* (blue).  $V_G = 0.3$  V,  $V_{DS} = -1.0$  V,  $n = 3$ ; (b) drain-source current for sterile LB broth and those containing several concentrations of *Pseudomonas fluorescens*:  $3.6 \times 10^{10}$ ,  $1.2 \times 10^6$ ,  $6.6 \times 10^8$  CFU/mL at  $V_G = 0.7$  V, 0.6 V, and 0.5 V. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

testing in LB broth (Fig. 5a). This direction of change in device operation indicates that the primary signal mechanism should be from cell gate attachment. Future studies on the primary switching mechanism should be performed to confirm this hypothesis. Using equations from Armbruster et al. (Armbruster and Pry 2008), the limit of detection (LOD) based on the standard deviations of the lowest bacteria concentration and sterile samples is calculated to be  $\sim 880$  CFU/mL for  $V_G = 0.6$  V, with a confidence level of 90%.

## 5. Summary and conclusions

In summary, the effects of nutrient broths as operating OECT media were studied for optimization of OECT-based bacterial sensing. Usage of nutrient broths as an operating media was found to result in a large shift in effective gate voltage while preserving device transconductance, which determines its sensitivity, when compared to operation in pure salt solutions. This shift in effective gate voltage allows the OECT to operate at its peak transconductance point at lower voltages. Therefore, OECT devices operate favorably in broth media rather than in salt electrolytes. Additionally, the lower operating point also increases the potential readout current due to an increase in  $I_{OP}/I_0$ . This broth induced shift is believed to be caused by redox reactions taking place between the gate electrode and yeast extracts and proteins in the media. Future studies will need to be done to confirm PEDOT:PSS redox reactions in broth media.

The operation of OECTs in an agar gel media was also studied, primarily through switching speed analysis. This study found that operation in a nutrient agar media resulted in much slower switching-off speeds compared to that in solution media. In general, the switching-on speeds were found to be slower than switching-off speeds.

In summary, bacteria detection without specific binding was demonstrated in LB broth using two methods. The first was the measurement of OECT source-drain current over a 4-h period with sterile and high bacterial concentration samples. The second method was evaluation of OECT source-drain current for fixed voltage parameters at varying concentrations of bacteria. In each case, presence of bacteria resulted in an increase in effective gate voltage. The calculated limit of detection achieved is  $\sim 880$  CFU/mL, comparable to the  $10^3$  CFU/mL of previous OECT bacteria detection utilizing antibody based specific capture (He et al., 2012). This demonstrates that unbound bacterial detection can successfully be achieved in complicated media without the aid of a capture agent. Future studies are planned to determine sensitivity in other non-broth media and for the detection of different strains in commercial environments.

## CRediT authorship contribution statement

**Eric Frantz:** Methodology, Investigation, Writing – original draft. **Jingchu Huang:** Methodology. **Daewoo Han:** Methodology, Writing – review & editing. **Andrew J. Steckl:** Conceptualization, Writing – review & editing.

## 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.

## Data availability

Data will be made available on request.

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## Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.biosx.2022.100268>.

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**The effect of nutrient broth media on PEDOT:PSS gated OECTs for whole-cell bacteria detection**

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**Supplemental Information**



Table S1 Examples of media in which OECTs have been operated.

<b>1<sup>st</sup> Author (PI)</b>	<b>Institution</b>	<b>Application</b>	<b>Operating Media</b>	<b>Year</b>
Preziosi	University Federico II	Blood characterization	Blood and plasma	2022
Méhes	Linköping Univ.	Bacteria monitoring	M9 buffer	2020
Wei (Lin)	Shenzhen Univ.	Cell monitoring	Polysaccharide medium	2017
Liao (Lin)	Hainan Univ.	Cell detection	Artificial seawater	2014
He (Yan)	Hong Kong Polytechnic	Bacteria detection	KCl	2012
Lin (Yan)	Hong Kong Polytechnic	Ion detection	Various salts	2010
Yang (Malliaras)	Cornell University	Enzymatic detection	PBS	2010

Table S2 Recipes for LB Miller and YT broth in 1L of water.

Broth/Ingredients	Tryptone (g/L)	Yeast Extract (g/L)	NaCl (M)
LB Miller	10	5	0.17
YT	16	10	0.085

Table S3 Comparison of key characteristics of OECTs using NaCl salt solutions, LB and YT broths and agars: peak transconductance and associated voltage, ratio of operating current to starting current.

Media	Peak $g_m$ (mS)	$V_{G,peak}$ (V)	$I_{OP}/I_0$
NaCl – 0.085M (n=4)	$2.83 \pm 0.44$	$0.62 \pm 0.02$	$0.20 \pm 0.03$
NaCl – 0.17M (n=4)	$3.30 \pm 0.92$	$0.72 \pm 0.05$	$0.33 \pm 0.02$
YT Broth (n=5)	$2.92 \pm 0.97$	$0.36 \pm 0.05$	$0.52 \pm 0.02$
LB Broth (n=5)	$3.04 \pm 1.14$	$0.28 \pm 0.04$	$0.59 \pm 0.02$
YT Agar (n=2)	$1.7 \pm 0.5$	$0.3 \pm 0.1$	$0.61 \pm 0.006$
LB Agar (n=2)	$1.5 \pm 0.4$	$0.3 \pm 0.0$	$0.59 \pm 0.002$

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