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A Novel Antibacterial Membrane Electrode Based on Bacterial Cellulose/Polyaniline/$AgNO_3$ Composite for Bio-Potential Signal Monitoring

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ABSTRACT We propose a flexible, dry, and antibacterial electrode with a low and stable skin electrode contact impedance for bio-potential signal monitoring. We fabricated a bacterial cellulose/polyaniline/$AgNO_3$ nanocomposite membrane (BC/PANI/$AgNO_3$) and used it for bio-potential signal monitoring. The bacterial cellulose (BC) provides a 3-D nanoporous network structure, and it was used as a substrate material in the BC/PANI/$AgNO_3$ nanocomposite membrane. Polyaniline (PANI) and $AgNO_3$, acting as conductive and antibacterial components, respectively, were polymerized and deposited on the surfaces of BC nanofibers to produce uniform thin film membrane with flexible, antibacterial, and conductive properties. Various measurements were conducted, in terms of antibacterial activity, skin electrode contact impedance, and qualitative analysis of ECG signal recordings. The BC/PANI/AgNO3 membrane revealed 100% antibacterial activities against both the *Staphylococcus aureus* and *Escherichia coli* bacteria. The skin electrode contact impedance of the proposed BC/PANI/$AgNO_3$ electrode is lower than that of the Ag/AgCl gel electrode, with the same active area. In addition, the electrocardiogram (ECG) signals acquired with the proposed electrodes have stable characteristic waveforms, and they are not contaminated by noise. The waveform fidelity of the BC/PANI/$AgNO_3$ membrane electrodes over 800 ECG cardiac cycles is 99.49%, and after the electrodes were worn for 24 hours, a fidelity of 98.40% was recorded over the same number of cardiac cycles. With the low and stable skin electrode contact impedance, the proposed dry BC/PANI/$AgNO_3$ membrane electrode provided high fidelity for ECG signal recordings, thus offering a potential approach for bio-potential signal monitoring. With the above benefits, the novel flexible and dry BC/PANI/$AgNO_3$ electrode has a significant antibacterial. Most of all, it is the first research to develop antibacterial in the electrode design.

INDEX TERMS Flexible and dry electrodes, bacterial cellulose/polyaniline/$AgNO_3$ nanocomposite membrane (BC/PANI/$AgNO_3$), antibacterial activity, low and stable contact impedance, bio-potential signal monitoring.

I. INTRODUCTION

Monitoring bio-potentials, such as electrocardiogram (ECG), electromyogram (EMG) and electroencephalogram (EEG), is an important step towards a better understanding of the pathological and physiological conditions in human.

To prevent diseases, wearable bio-potential systems monitoring the physiology of patients and personal healthcare have attracted lots of attention. This is essential for long-term monitoring or to obtain 24-hour continuous recordings in the case of ECG, EEG, and various other bio-signals.

However, the electrodes used for bio-potential acquisition system directly affect the quality of the bio-potential signal. The development of a flexible and dry electrode with low and stable skin electrode contact impedance for long-term bio-potential monitoring has being an important area of research.

Due to reliability, the traditional silver/silver chloride (Ag/AgCl) gel electrodes are widely used in clinical bio-potential monitoring. Typically, these traditional Ag/AgCl gel electrodes transforms ionic currents in the body to electronic currents by two interfaces: electrode-electrolyte interface and electrolyte-skin interface [1]–[3]. Therefore, the conductive gel is required for efficient operation of the traditional Ag/AgCl electrode. However, conductive gel could cause itchiness, reddening, and swelling. These can cause the bacteria to get trapped on the skin. So, traditional Ag/AgCl gel electrodes have a risk of skin irritation and are not suitable for long-term monitoring [4].

On the contrary, the dry electrode without explicit conductive gel could become alternative candidate for ambulatory and continuous registration of bio-potentials if conformal contact to the skin can be ensured such that the contact impedance is lowered and stable [5]. In order to obtain low skin-electrode contact impedance, various micro-needle electrodes have been proposed [6]–[8]. For instance, Salvo *et al.* [9] proposed a prototype of micro-needle dry electrode based on photopolymer for ECG/EEG recording. However, micro-needle dry electrode is an invasive measurement in terms of its working principle, and micro-needle electrode is easy to break in the skin owing to its lack of sufficient strength [10], [11]. Zhang *et al.* [11] designed stiff and sharp silicon microneedles to penetrate the stratum corneum layer. Also, based on the advances in conductive textile materials, a number of recent studies have proposed the use textile electrodes for biopotential signal acquisition. For example, Tong *et al.* [12] developed a nanofiber web textile dry electrodes for long-term biopotential signal recording. Bahareh *et al.* [13] applied pressure technique to minimize skin electrode contact impedance. Though the above proposed methods could effectively handle the drawbacks of micro-needle dry electrode, but electroconductive textile-based dry electrodes often produce more noise especially in low frequency bands (0.00 Hz - 0.67 Hz) of the recordings [14]–[18]. It should also be noted that textile-based electrodes which are knitted, woven, and embroidered into clothes tend to exhibit different electrical properties upon stretching and eventually result in discomfort during persistent motion [19].

Polydimethylsiloxane (PDMS) has been widely used in dry electrodes fabrication for its good elastic properties, flexibility, biocompatibility and optical transparency [20]. To date, the polymer-based dry electrodes are sub-categorized into carbon-based electrodes, printed electrodes, and metal coating electrodes. Jung *et al.* [21] fabricated a carbon nanotube (CNT)/ PDMS composite-based dry ECG electrode. Kim *et al.* [22] incorporated CNT and grapheme into elastomeric matrix. However, the dispersion of CNT or grapheme into PDMS are challenging. Variants of patterning conductive media directly on a PDMS substrate have attracted much interest [23]–[26]. Among these studies, direct imprinting of silver nanoparticles and direct ink-writing have been reported as simple and cost-effective methods. However, the patterning methods should leave no conductive residual layer between conductive patterns, to avoid short circuit. Well-developed functional inks is also a key point in achieving high electrical performance. Even though, PDMS material have a good contact with skin due to flexibility of the used polymer, but they lack the ability of reducing the contact impedance at low frequencies and are still sensible to motion artifacts [27]. In order to guarantee a very stable interface with the skin for long-term monitoring; Kim and Kim [28] used PDMS stamp to attach electrodes to skin. However, the PDMS stamp does not perform well in long-term monitoring [29]. Bae [30] and Lee group [31] fabricated microstructure in PDMS stamp respectively. These approaches have proven to be useful, but they require expensive and sophisticated processes, and the utility of these methods is limited. Recently, some studies [19], [32]–[35] have tried to use sweat to reduce the contact impedance of dry electrode. But the electrode based on these new methods have not been widely adopted due to poor performance. To the best of our knowledge, no research has considered the bacterial infections on skin when the skin is breached or under moist occlusive conditions, especially during long-term bio-potential monitoring. Given the large number of inherent defense mechanisms, the skin is a poor media for bacteria. However, when altered or breached or under moist occlusive conditions, the skin can support the growth of both commensal and pathogenic bacteria which protect the host from pathogenic bacteria. It is worthy to note that environmental and local factors as well as organism adherence and virulence are intricately related to cutaneous infection that may severely affect the skin [37].

Due to stable nanofiber network, high chemical purity, high degree of polymerization, excellent biocompatibility and biodegradability, bacterial cellulose (BC) is one great matrix and substrate material. In addition, BC is flexible, it has good mechanical properties, and it can easily change its size and shape, accordingly, in response to environmental stimuli. Therefore, BC open up the important and rapidly expanding fields of personal care, medicine, and life sciences [46]. When nanocellulose is combined with conductive materials, it has potential to monitor bio-potential signal. Polyaniline (PANI) is one promising conductive polymer, because of its ease of synthesis, low cost monomer, tunable properties, and high environmental stability [36]. Recently, bacterial cellulose and polyaniline nanocomposite membrane (BC/PANI) have been developed [53], [54]. PANI was polymerized and deposited as an adhering layer on the surfaces of the nanocellulose fibers. This forms a uniform thin film conductive material, and its conductivity can reach 5.2×10^{-3}S cm-1 [38]. Therefore, BC/PANI will yield potential designs and fabrication of long-term bio-potential monitoring if antibacterial property is developed.

In this paper, we developed bacterial cellulose/polyaniline/$AgNO_3$ composite membrane (BC/PANI/$AgNO_3$), and used it in bio-potential signal monitoring. It should be noted that this study is the first to utilize BC/PANI concept and consider antibiosis for electrode design and fabrication. The proposed flexible and dry electrode was evaluated based on skin electrode contact impedance, waveform fidelity, and antibacterial measurements, and compared with traditional Ag/Cl gel electrode.

II. MATERIALS AND FABRICATION

A. DESIGN

Note that the average diameter of pure BC nanofiber is about 40 nm, and the length ranges from micrometers up to dozens of micrometers [46], [32]. This yields a three-dimensional polymeric network, and has a large contact area as the electrode material. As a conductive polymer material, PANI forms conductive network by in situ polymerization on the BC fiber. BC, rich in hydroxyl group, has good affinity with Polyaniline. Hydroxyl groups of BC interact with amine groups of aniline to form hydrogen bond which could ensure the uniform and stable distribution of aniline on the surface of BC nanofibers. Therefore, PANI facilitates the development of BC with high conductive property. Antimicroia $AgNO_3$ make the nanocomposite membrane antibacterial.

B. MATERIALS

BC membrane was provided by Hainan Yida Food Co., Ltd (China). Sodium periodate ($NaIO_4$), sodium hydrogen sulfite ($NaHSO_3$), aniline, and $AgNO_3$ were purchased from Sinopharm Chemical Reagent Co., Ltd (China). Traditional Ag/AgCl gel electrodes (red dot, 2238) were purchased from 3M Co., Ltd (USA).

C. FABRICATION

The BC were placed in a beaker containing a mixture of 50 mL hydrochloride (HCl) solution (1.0 mol/L) and 0.025 mol dissolved aniline monomers. The BC membranes were fully immerged in the above mentioned solution for 24 hours, then oxidant of 0.2 mol/L ammonium peroxide sulfate (APS) was added, dropwise, into the solution that waggles in 200 rpm. The mixture was kept in an ice bath for 90 min with unremitting stir. After the experiment, the obtained composite membranes were brought out and washed with 75% (v/v) ethyl alcohol. Afterwards, the membranes were washed thoroughly using deionized water in order to remove the byproducts and remnant reagents.

After the oxidant of APS was dripped into the aniline HCl solution, the aniline monomer would, in situ, polymerize to form polyaniline in the network of BC and the ivory membranes changed to dark green ones, then the BC/PANI composite electrolyte membranes have been synthesized. The BC/PANI composite electrolyte membrane was immersed in 0.1 moL/L $AgNO_3$ solution for 5 minutes and then, Ag^+ was deposited in the porous structure of BC.

III. MEASUREMENTS

A. ANTI-BACTERIAL MEASUREMENT

Antibacterial activities of the proposed electrode and that of the traditional Ag/AgCl gel electrode were tested against two bacteria including Staphylococcus aureus (*S.aureus*) and Escherichia coli (*E.coli*), typical representatives of Gram+ and Gram- bacteria, which are considered as common infectious microorganisms [39]. We developed antibacterial ring experiment and bacterial colony experiment to evaluate the antibacterial activities of the proposed BC/PANI/$AgNO_3$ membrane electrode.

A sample of the proposed membrane electrode that assumes a disc-like shape with 8 mm diameter was designed. Similarly, a sample of the traditional Ag/AgCl electrode with the same size and shape was equally prepared in aseptic conditions.

Staphylococcus aureus and Escherichia coli concentration of 105 CFU/mL were diluted by 1:100 and inoculated on the Luria-Bertani (LB) agar plate. Then, the swatches were placed at the center of the agar plate and inoculated at 37°C for 24 hours. Bacteriostasis ring of the proposed electrode and gel in traditional Ag/AgCl gel electrode were measured. Bacterial suspension with 60uL in concentration of 105 CFU/mL was dripped in another series of the samples and were incubated at 37°C for 24 hours. Then, the swatches were washed in 1mL LB culture solution and placed in falcon tubes. The tubes were ultrasonic for 5 minutes so as to make the bacteria fully detached from surface of the samples. Bacterial suspension of 2mL in falcon tubes was chosen; and these were inoculated on the Luria-Bertani (LB) agar plate at 37°C for 24 hours. Finally, the colonies were counted and concentration of bacteria in each sample was determined. The reduction rate in the number of test microorganisms was calculated as:

$$BR(\%) = 100 \times (A - B)/A \tag{1}$$

where BR is the percentage reduction rate; and A, B are the number of microorganisms recovered from the inoculated proposed electrode and gel in traditional Ag/AgCl electrode after 24 hours, respectively.

B. SKIN ELECTRODE CONTACT IMPEDANCE MEASUREMENT

The skin electrode contact impedance was measured with Electrochemical Analyzer/Workstation (700E, CH Instruments Inc., America) at frequencies ranging from 1 to 10 kHz. For each measurement, a pair of BC/PANI/$AgNO_3$ membrane electrodes were placed on the forearm of the three healthy male subjects recruited in the study. And the electrodes were placed 5cm apart from each other. The skin electrode contact impedance of the traditional Ag/Cl gel electrodes was also measured in the same anatomical area. Prior to each measurement, cleaning with alcohol, shaving or scrubbing of the skin surface area were not done.

Before the experiment began, all the recruited subjects were clearly informed about the objective of the study.

Afterwards, they all agreed and provided written informed consent as well as permission for the publication of their data for scientific and educational purposes. The entire protocol of this study was approved by the Institutional Review Board of Shenzhen Institutes of Advanced Technology, Chinese Academy of Sciences, China.

C. BIO-POTENTIAL MEASUREMENT

The ECG100C module provided by BIOPAC Acquisition systems, Inc. (Goleta, CA, USA) were used to acquire ECG signals. For comparison purposes, ECG signals were recorded from a male healthy subject (23 years old) using traditional Ag/Cl gel electrode and proposed electrode simultaneously. The traditional gel Ag/Cl electrodes have an active circular area with a diameter of 16mm and the diameter of BC/PANI/$AgNO_3$ electrodes also have the same size with the Ag/Cl electrodes.

BIOPAC acquisition systems provide a frequency response between 0.5 to 35 Hz and sampling frequency is 1000Hz. We recorded two ECG signals for 12 minutes using a pair of the proposed electrodes and a pair of the traditional Ag/AgCl gel electrodes simultaneously based on the electrode configuration shown in Figure 6. The signal contain 800 cardiac cycles and are used for bio-potential analyses.

D. BIO-POTENTIAL ANALYSES

1) PEAK-TO-PEAK AMPLITUDE REDUCTION QUANTIFICATION

According to [41], peak-to-peak amplitude reduction quantification is one of the important measurement parameters for bio-potential signals. The signal acquired by the proposed BC/PANI/$AgNO_3$ membrane electrode contained 800 cardiac cycles of peak-to-peak amplitudes. Similarly, the second pair of the traditional Ag/AgCl gel electrodes (described in Section III, Part C) was used to quantify the amplitude reduction of the ECG signals alongside the co-located proposed electrodes. The R-wave peak in each cardiac cycle was detected according to the method in described [42], [43].

2) WAVEFORM FIDELITY

Here, we utilized 800 cardiac cycles recorded with a pairs of the proposed electrodes and the traditional Ag/AgCl gel electrodes simultaneously (as described in Section III, Part C) to compute the waveform fidelity. It is noteworthy that the 800 cardiac cycles were segmented and from the segments, an averaged ECG signal over one cardiac cycle was computed. We scaled the amplitudes of the averaged ECG cycle, as measured by the proposed electrode, to match their R-wave amplitudes uniform with that of the average ECG signal measured using the traditional Ag/AgCl gel electrode. The ECG signal measured using the traditional Ag/AgCl gel electrodes were considered as the reference signal. Thus, the differences between the reference signal measured by the traditional Ag/AgCl gel electrode and that measured by the proposed electrodes were computed and denoted as 'error signal'. The 'waveform fidelity' was found to be 100% less than the percentage of the magnitude of the relative error with respect to the true signal [12]. This is obtained using Eq. 2.

$$WF(\%) = 100 \times \left\{ 1 - \left| \frac{(X_{Ag/AgCl} - X_{test})}{X_{Ag/AgCl}} \right| \right\} \tag{2}$$

where $X_{Ag/AgCl}$ is the averaged ECG cycle trace recorded by Ag/AgCl electrode, and X_{test} is the averaged ECG cycle trace recorded by proposed electrode.

3) PSD AND COHERENCE ANALYSES

We choose two ECG signals recorded using the proposed and traditional Ag/AgCl gel electrodes (as described in Section III, Part C) to carry out the power spectral density (PSD) and coherence analysis.

E. BIO-POTENTIAL MEASUREMENT OF LONG-TERM WEAR

A pair of proposed BC/PANI/$AgNO_3$ and Ag/AgCl gel electrodes were used for bio-potential signal measurement during long-term wearing. The Ag/AgCl gel electrode is used as reference. ECG signals were recorded for a period of 15 minutes using both the proposed and Ag/AgCl gel electrodes, simultaneously. Then, another set of ECG recordings were made for 15 minutes with the same experimental setup after the proposed BC/PANI/$AgNO_3$ electrodes were worn for 24 hours.

IV. RESULTS AND DISCUSSION

A. ELECTRODE FABRICATION

To evaluate the morphological characteristic of the fabricated BC/PANI membrane electrode, scanning electron microscope (SEM) images were obtained. The porous structure of the fiber network was clearly visible through the SEM. Figure 1 (a) and (b) show that the FE-SEM of pure BC and BC/PANI/AgNO3 nanocomposite membranes respectively. It is obvious that the BC fiber was fully encased with PANI nanoparticles to form a core-shell structured morphology with polyaniline coated on the BC fibers. Due to strong hydrogen bonding formed between PANI layers and BC fibers [44], [45], polyaniline was coated on the BC fibers. Note that the average diameter of pure BC nanofiber is about 40 nm, and the length ranges from micrometers up to dozens of micrometers [46], [32]. This porous structure could increase contact area between the skin and the electrode.

The fabricated BC/PANI/$AgNO_3$ membrane electrode exhibited flexible characteristics, as shown in Figure 2 (a). In order to compare performance of the proposed and traditional Ag/AgCl electrodes, in terms of skin-electrode contact impedance and physical activity such as: power spectral density, waveform fidelity, the fabricated electrode membrane are diced into circulars shapes which have same size with the active area of Ag/AgCl electrode. The diced electrode is in diameter of 16 mm, and the active area of traditional Ag/AgCl electrode (red dot 2238, 3M) has same diameter value. The diced BC/PANI/$AgNO_3$ membrane electrode was attached on

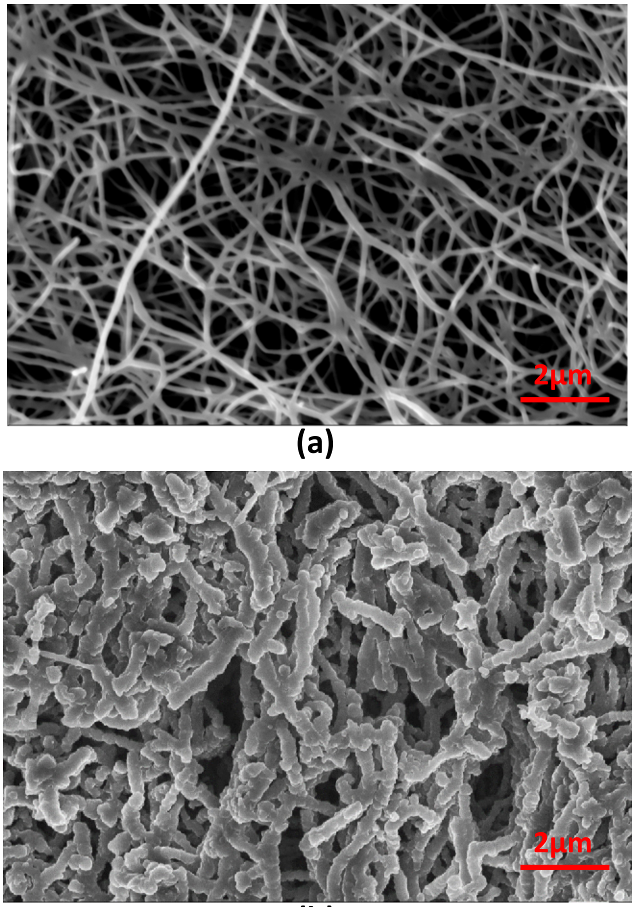

FIGURE 1. (a)FE-SEM images of BC membranes. (b)FE-SEM images of BC/PANI/$AgNO_3$ nanocomposite membranes. Scale bar = 2000nm.

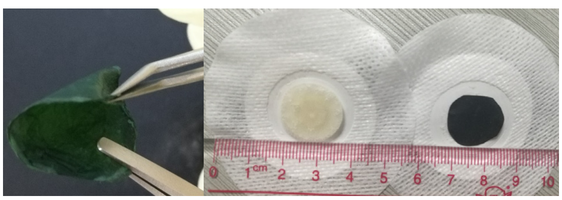

FIGURE 2. Image of the flexible and dry BC/PANI/$AgNO_3$ membrane electrode. (a)The BC/PANI/$AgNO_3$ membrane was bent by tweezers. (b) Traditional Ag/AgCl gel electrode which active area is in diameter 16 mm and the BC/PANI/$AgNO_3$ electrode in diameter 16 mm. In the proposed electrode, the gel from Ag/AgCl electrode was replaced with the BC/PANI/$AgNO_3$ membrane.

one Ag/AgCl tape with metallic snap fastener on the other side, as shown in Figure 2(b). The design make the proposed electrode a direct connection with standard patient cables.

B. MEASUREMENT OF ANTIBACTERIAL

Antibacterial activities of BC/PANI/$AgNO_3$ nanocomposite membrane and gel from the traditional Ag/AgCl electrode were investigated against two kinds of bacteria (i.e. Gram+ (*S.aureus*) and Gram-(*E.coli*)). Antibacterial ring experiment and bacterial colony experiment were carried out respectively. The results of bacteriostatic ring test (Figure 3) showed that the proposed SBC/PANI membrane has antibacterial activity against *E.coli* and *S.aureus*, especially on *S.aureus*. However, the traditional Ag/AgCl gel electrode has no obvious antibacterial effect on the two kinds of bacteria.

The further colony count test showed that the antibacterial resistance rate of proposed BC/PANI/$AgNO_3$ membrane against *E.coli* and *S.aureus* reached 100%, while the Ag/AgCl gel group had no antibacterial effect. In other words, the results show that there is no antimicrobial effect on gel from traditional Ag/AgCl gel electrode.

C. SKIN AND ELECTRODE CONTACT IMPEDANCE CHARACTERIZATION

We measured and compared the skin-electrode contact impedance of the proposed electrode and traditional Ag/AgCl (red dot 2238, 3M) electrode on three volunteer's forearm. Figure 5 show the results of skin electrode contact impedance presented as the mean ± standard deviation versus frequency, 1-10 kHz. Compared to the Ag/AgCl gel electrode, the proposed electrode has lower skin electrode contact impedance at frequency range 1-100Hz. As low frequency range (<100Hz) is especially beneficial for bio-potential monitoring, the skin electrode contact impedance at 10Hz is further compared with electrodes in researches [11], [33], [35], [47]–[51].

Table 1 summarized the skin electrode contact impedance of the proposed electrode and other classical electrodes as reported in literature. The mean skin electrode contact impedance of the proposed electrode is 51 KOhm without any preprocessing. The contact impedance value of the proposed electrode is significantly lower than the impedance values in [33], [47]–[51] and comparable with the impedance values of electrodes in [35] and [11]. However, the original impedance of the self-wetting electrode proposed in [35] is 240 KOhm. After dropping a little saline to moisten the surface of the self-wetting electrode, the skin electrode contact impedance of self-wetting electrode reduced to 66 KOhm. Micro needle electrode [11] is invasive.

TABLE 1. Contact impedance of different electrodes at 10HZ.

Electrode	Contact Impedance (KOhm)	Morphology structure	Interface Material
1[47]	135	Micro pillars	Ag
2[48]	150	Micro domes	Ag
3[49]	400	Micro domes	Ni/Pt
4[50]	500	Serpentine shape	CNT
5[33]	700	Micro pillars	CNT
6[51]	800	Macro-convex	Ti/Au
7[35]	66	Porous fiber	PEDOT/PSS
8[11]	61	Micro needle	PEDOT/PSS
This paper	51	Porous fiber	PANI

The achieved results revealed the capability of the proposed electrode to work without the electrolyte. It is obvious that the proposed electrode could provide valuable interface

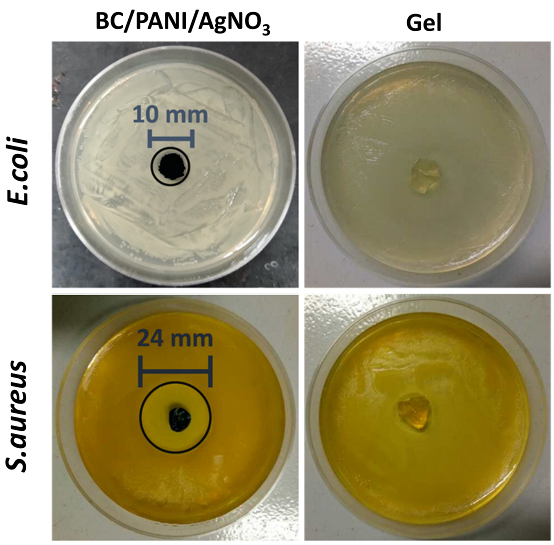

FIGURE 3. Antibacterial rings against *E.coli*and *S.aureus* for BC/PANI/$AgNO_3$ membrane sample and gel from Ag/AgCl electrode.

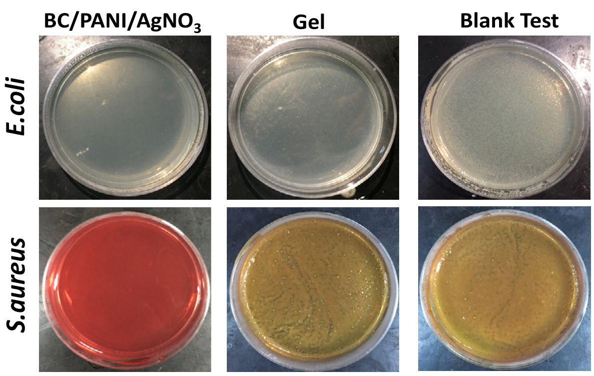

FIGURE 4. Bacterial colony experiment against *E.coli* and *S.aureus* for BC/PANI/$AgNO_3$ membrane sample and gel from Ag/AgCl electrode.

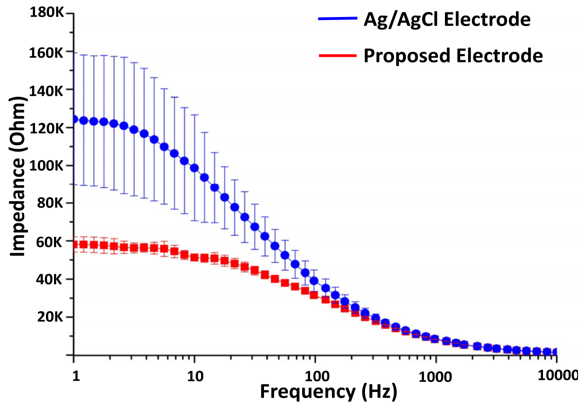

FIGURE 5. Skin and electrode contact impedance comparison of the proposed SBC/PANI electrodes and traditional Ag/Cl gel electrode.

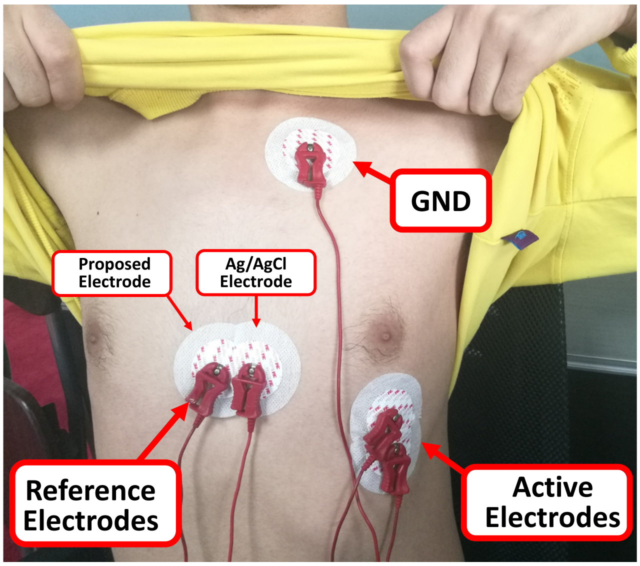

FIGURE 6. The electrode configuration utilized for ECG signal acquisition. The configuration is consist of a pair the proposed BC/PANI/$AgNO_3$ electrodes, a pair of the traditional Ag/AgCl gel electrode as well as a ground (GND) electrode. Kindly note that the configuration also contains reference electrodes corresponding to a pair of active electrodes for both the proposed and traditional Ag/AgCl electrodes. The proposed and traditional Ag/AgCl electrodes have been shown in Figure 2.

with the skin. The 3D porous structure of BC, which improves the contact area between the skin and electrode, and mixed conductivity offered by PANI, electronic conductivity and ionic conductivity, contributed to this.

D. BIO-POTENTIAL MEASUREMENT

1) ECG WAVEFORMS CONTRAST

The fabricated BC/PANI/$AgNO_3$ electrode and Ag/AgCl gel electrode as shown in Figure 2(b), were used to record ECG signals simultaneously according the electrodes configuration described in Figure 6. The ground (GND) electrode, a single reference and a single active electrode would collectively produce one channel ECG recording, and this configuration mimics the traditional Lead II ECG described in [40] and [41]. The GND electrode was placed on the xiphoid process and the reference electrode is placed on center of the manubrium. Meanwhile, the active electrode is placed under the left pectoral near the V6 electrode position. And one traditional Ag/Cl gel electrode was used as ground electrode. Kindly note that the configuration also contains reference electrodes corresponding to a pair of active electrodes for both the proposed and traditional Ag/AgCl electrodes.

Figure 7 shows a representative example of ECG recorded using a pair of the proposed electrodes and a pair of the traditional Ag/AgCl gel electrodes simultaneously without any preprocessing. Figure 8 shows an example of ECG cardiac cycle. As it could be seen from Figures 7 and 8, compared to traditional Ag/AgCl gel electrodes, the ECG signals acquired with the proposed electrodes have stable characteristic waveforms and were not contaminated by noise.

The porous structure and flexible mechanical property of BC make the BC/PANI/$AgNO_3$ electrode easily change its size and shape in response to environmental stimuli. Therefore, the proposed electrode could fit well to the skin which

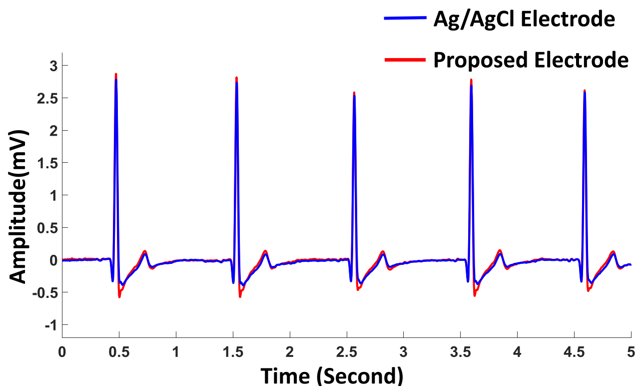

FIGURE 7. One ECG Signal example recorded using proposed electrodes and traditional Ag/AgCl electrodes simultaneously according electrode configuration shown in Figure 6.

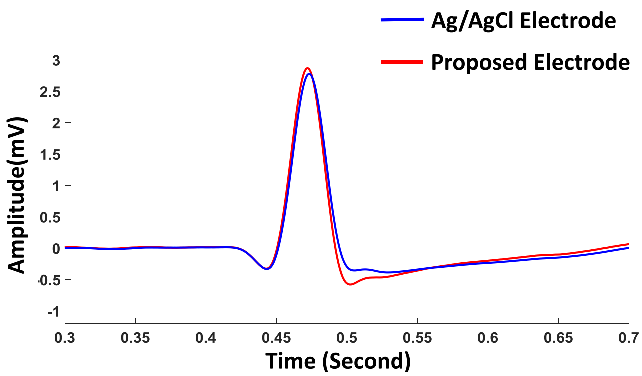

FIGURE 8. One ECG cardiac cycle recorded using proposed electrode and traditional Ag/AgCl electrodes simultaneously according electrode configuration shown in Figure 6.

reduce the noise contamination and baseline drift effectively in ECG signal acquisition. However, the signals recorded with the textile electrodes, proposed by Chen *et al.* [52], exhibit some variation in waveforms as well as amplitudes and are more contaminated by noise. Therefore, the proposed BC/PANI/$AgNO_3$ electrodes is able to acquire ECG signals that are highly correlated with those from traditional Ag/AgCl gel electrodes even feature waveforms. Note that for fair comparison between the ECG recordings obtained with the proposed electrode and the traditional Ag/AgCl gel electrodes, both electrodes were placed as close as possible as shown in Figure 6.This method minimizes the waveform difference caused by position difference between measurement electrodes.

2) PEAK-TO-PEAK AMPLITUDE QUANTIFICATION

Figure 9 shows a boxplot with the results obtained for the peak-to-peak amplitude using both the proposed BC/PANI/$AgNO_3$ and Ag/AgCl electrodes over 800 cardiac cycles. It is clearly visible that the proposed BC/PANI/$AgNO_3$ electrode and Ag/AgCl gel electrode have a similar performance in amplitudes obtained over 800 cardiac cycles. In fact, compared with Ag/AgCl gel electrode, the proposed BC/PANI/$AgNO_3$ electrode present a slightly lower dispersion with similar median values.

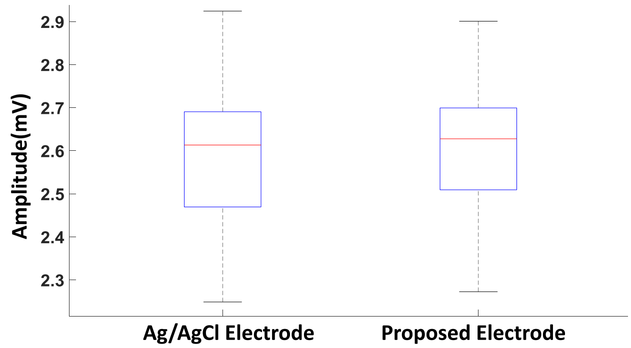

FIGURE 9. Peak-to-peak amplitude over 800 cardiac cycles recorded using proposed electrodes and traditional Ag/AgCl electrodes simultaneously according electrode configuration shown in Figure 1.

3) WAVEFORM FIDELITY

By using the traditional gel Ag/AgCl electrode as reference, the waveform fidelity value of the proposed electrode was computed. We measured the average ECG waveforms over 800 cardiac cycles using the proposed electrodes and traditional gel Ag/AgCl electrode. In order to compute the waveform fidelity, the two averaged cardiac cycle waveforms have been scaled to make them have the same R-wave amplitudes, as shown in Figure 10.

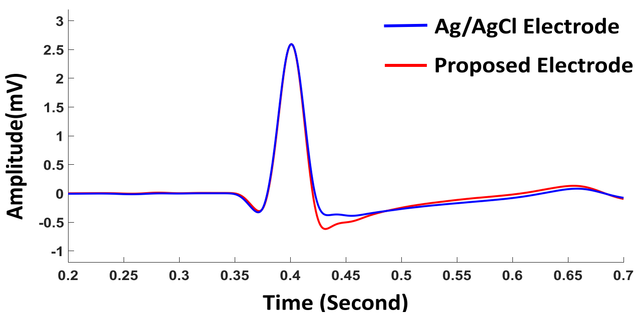

FIGURE 10. Averaged ECG waveforms over 800 cardiac cycles using proposed electrode and traditional gel Ag/AgCl electrode. The amplitudes of two averaged ECG waveforms have been scaled to make their R-wave amplitudes.

The computed waveform fidelity value is 99.49%. Compare to the nanofiber web textile dry electrodes proposed by Tong *et al.* [12], the highest achieved waveform fidelity was 95%. It is obvious that the proposed electrodes have significantly higher performance in gathering biopotential recording compared to nanofiber flexible textile electrode.

4) PSD CONTRAST AND COHERENCE ANALYSES

Figure 11 and Figure 12 show the power spectral density (PSD) contrast and the coherence analyses, respectively, between the ECG signals which are obtained with the proposed electrode and the traditional Ag/AgCl gel electrodes. As shown in Figure 11, the PSD of the

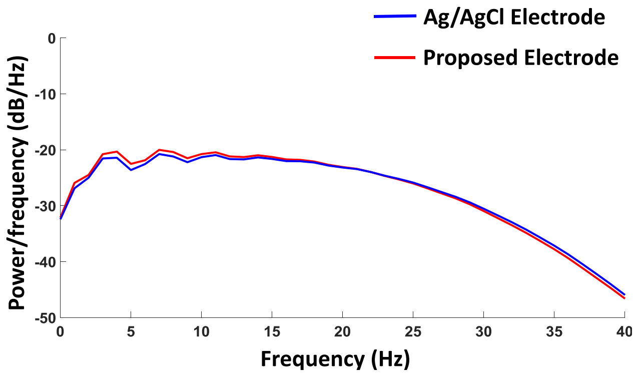

FIGURE 11. **PSD contrast of recorded ECG signals used proposed electrode and traditional Ag/AgCl electrodes.**

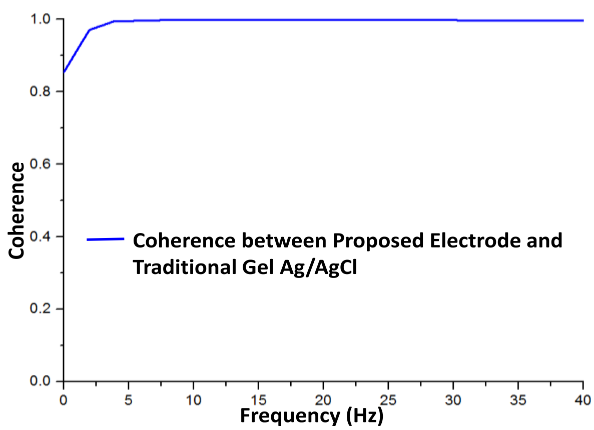

FIGURE 12. **Coherence analyses of recorded ECG signals used proposed electrode and traditional Ag/AgCl electrode.**

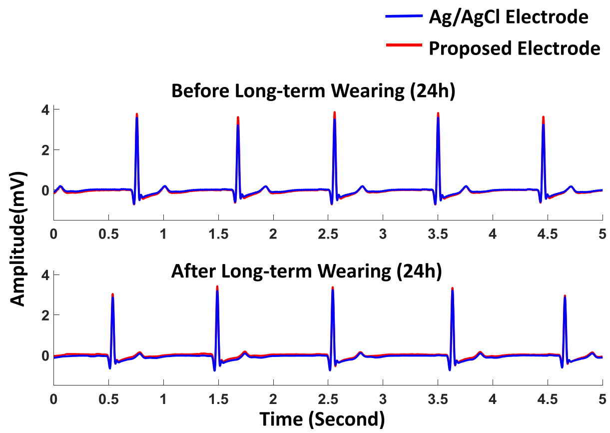

FIGURE 13. **One ECG signal example used by proposed BC/PANI/$AgNO_3$ electrode and traditional Ag/AgCl gel electrode before and after long-term wearing (24 hours).**

ECG signals obtained by proposed electrode and Ag/AgCl electrode is quite similar at frequencies (0.5Hz-35Hz), BIOPAC system provided, which is among the main frequency of ECG. Besides, the coherence is approximately equal to 1 in the measured frequencies (0.5Hz-35Hz). Remarkably, the result for proposed BC/PANI/$AgNO_3$ electrode is comparable to that of Ag/AgCl gel electrodes, meaning that the proposed electrolyte-free approach is able to deal with ECG signal acquisition.

E. BIO-POTENTIAL MEASUREMENT OF LONG-TERM

The ECG signals were recorded using the proposed BC/PANI/$AgNO_3$ and the traditional Ag/AgCl gel electrodes before and after 24 hours wearing. The signals obtained are plotted in Figure 13 without any preprocessing. Before 24 hours long-term wearing, the ECG signal were recorded for 15 minutes using the proposed electrode and traditional Ag/AgCl electrode in which 800 ECG cardiac cycles were selected to compute the waveform fidelity. The value of waveform fidelity is 99.49%. After the proposed electrodes were worn for 24 hours, the computed waveform fidelity over 800 ECG cardiac cycles is 98.40%. As shown in Figure 13, the ECG signal recorded using the proposed BC/PANI/$AgNO_3$ electrode still have stable characteristic waveforms, and were not contaminated by noise. We found that, there is almost no difference in the waveform fidelity value and waveforms contrast before and after 24 hours long-term wearing. The strong hydrogen bonding between the PANI layers coated on the BC fibers steadily made the electrode have stable electrical characteristics and stable skin-electrode contact impedance. The proposed electrode has stable performance in long-term bio-potential monitoring. Therefore, the proposed BC/PANI/$AgNO_3$ electrode could be suitable for long-term monitoring in practical applications.

V. CONCLUSION

In this study, we successfully fabricated BC/PANI/$AgNO_3$ nanocomposite electrode and used it for bio-potential monitoring. BC is used as substrate material and PANI forms the conductive network by polymerization and deposit on the surfaces of the BC nanocellulose fibers. Compared with Ag/AgCl gel electrode, the proposed BC/PANI/$AgNO_3$ electrode has lower skin-electrode contact impedance and provides valuable interface with the skin during ECG signal acquisition. The waveform fidelity of the BC/PANI/$AgNO_3$ membrane electrodes is 99.49% and after the electrodes were worn for 24 hours, a fidelity of 98.40% was recorded over the same number of cardiac cycles. The spectral content of BC/PANI/$AgNO_3$ electrode and Ag/AgCl gel electrode is quite similar with a coherence that is approximately equal to 1. The above assessments indicate that the proposed BC/PANI/$AgNO_3$ electrode has a reliable and stable behavior during ECG signal recording, which is comparable or even better than that of traditional Ag/AgCl gel electrode. In addition, the proposed electrode achieved 100% antibacterial activities which is a remarkable feature in the design process of the electrodes. Lastly, the proposed BC/PANI/$AgNO_3$ electrode could potentially facilitate practical development of long-term health care systems.

It should be noted that the thickness of the electrode in the current study range between 50 $\sim$ 100μm. Therefore, it could be easily torn when the electrode is reused. One the other hand, it is very suitable for lightweight wireless acquisition system in long-term ECG, eye movement (EOG) and facial muscle activity (EMG) monitoring.

REFERENCES

[1] E. McAdams, "Biomedical electrodes for biopotential monitoring and electrostimulation," in *Bio-Medical CMOS ICs*. New York, NY, USA: Springer-Verlag, 2011, pp. 31–124.

[2] M. Oehler, V. Ling, K. Melhorn, and M. Schilling, "A multichannel portable ECG system with capacitive sensors," *Physiol. Meas.*, vol. 29, no. 7, pp. 783–793, 2008.

[3] K.-F. Wu and Y.-T. Zhang, "Contactless and continuous monitoring of heart electric activities through clothes on a sleeping bed," in *Proc. 5th Int. Conf. Inf. Technol. Appl. Biomed.*, May 2008, pp. 282–285.

[4] R. J. Prance *et al.*, "An ultra-low-noise electrical-potential probe for human-body scanning," *Meas. Sci. Technol.*, vol. 11, no. 3, pp. 291–297, 2000.

[5] N. Meziane, J. G. Webster, M. Attari, and A. J. Nimunkar, "Dry electrodes for electrocardiography," *Physiol. Meas.*, vol. 34, no. 9, pp. R47–R69, 2013.

[6] W. C. Ng *et al.*, "Micro-spike EEG electrode and the vacuum-casting technology for mass production," *J. Mater, Process. Technol.*, vol. 209., no. 9, pp. 4434–4438, 2009.

[7] N. S. Dias, J. P. Carmo, A. F. da Silva, P. M. Mendes, and J. H. Correia, "New dry electrodes based on iridium oxide (IrO) for non-invasive biopotential recordings and stimulation," *Sens. Actuators A, Phys.*, vol. 164, nos. 1–2, pp. 28–34, 2010.

[8] L. M. Yu, F. E. H. Tay, D. G. Guo, L. Xu, and K. L. Yap, "A microfabricated electrode with hollow microneedles for ECG measurement," *Sens. Actuators A, Phys.*, vol. 151, no. 1, pp. 17–22, Apr. 2009.

[9] P. Salvo, R. Raedt, E. Carrette, D. Schaubroeck, J. Vanfleteren, and L. Cardon, "A 3D printed dry electrode for ECG/EEG recording," *Sens. Actuators A, Phys.*, vol. 174, pp. 96–102, Feb. 2012.

[10] C.-Y. Chen, C.-L. Chang, T.-F. Chien, and C.-H. Luo, "Flexible PDMS electrode for one-point wearable wireless bio-potential acquisition," *Sens. Actuators A, Phys.*, vol. 203, pp. 20–28, Dec. 2013.

[11] H. Zhang *et al.*, "A motion interference-insensitive flexible dry electrode," *IEEE Trans. Biomed. Eng.*, vol. 63, no. 6, pp. 1136–1144, Jun. 2016.

[12] T. I. Oh *et al.*, "Nanofiber Web textile dry electrodes for long-term biopotential recording," *IEEE Trans. Biomed. Circuits Syst.*, vol. 7, no. 2, pp. 204–211, Apr. 2013.

[13] B. Taji, S. Shirmohammadi, V. Groza, and I. Batkin, "Impact of skin–electrode interface on electrocardiogram measurements using conductive textile electrodes," *IEEE Trans. Instrum. Meas.*, vol. 63, no. 6, pp. 1412–1422, Jun. 2014.

[14] V. Marozas, A. Petrenas, S. Daukantas, and A. Lukosevicius, "A comparison of conductive textile-based and silver/silver chloride gel electrodes in exercise electrocardiogram recordings," *J. Electrocardiol.*, vol. 44, no. 2, pp. 189–194, Mar./Apr. 2011.

[15] R. Paradiso, G. Loriga, and N. Taccini, "A wearable health care system based on knitted integrated sensors," *IEEE Trans. Inf. Technol. Biomed.*, vol. 9, no. 3, pp. 337–344, Sep. 2005.

[16] L. Beckmann *et al.*, "Characterization of textile electrodes and conductors using standardized measurement setups," *Physiol. Meas.*, vol. 31, no. 2, pp. 233–247, Feb. 2010.

[17] M. M. Puurtinen, S. M. Komulainen, P. K. Kauppinen, J. A. V. Malmivuo, and J. A. K. Hyttinen, "Measurement of noise and impedance of dry and wet textile electrodes, and textile electrodes with hydrogel," in *Proc. IEEE 28th Annu. Int. Conf. Eng. Med. Biol. Soc.*, Aug./Sep. 2006, pp. 6012–6015.

[18] J. S. Lee, J. Heo, W. K. Lee, Y. G. Lim, Y. H. Kim, and K. S. Park, "Flexible capacitive electrodes for minimizing motion artifacts in ambulatory electrocardiograms," *Sensors*, vol. 14, no. 8, pp. 14732–14743, 2014.

[19] M. A. Yokus and J. S. Jur, "Fabric-based wearable dry electrodes for body surface biopotential recording," *IEEE Trans. Biomed. Eng.*, vol. 63, no. 2, pp. 423–430, Feb. 2016.

[20] C.-T. Lin, L.-D. Liao, Y.-H. Liu, I.-J. Wang, B.-S. Lin, and J.-Y. Chang, "Novel dry polymer foam electrodes for long-term EEG measurement," *IEEE Trans. Biomed. Eng.*, vol. 58, no. 5, pp. 1200–1207, May 2011.

[21] H.-C. Jung *et al.*, "CNT/PDMS composite flexible dry electrodes for long-term ECG monitoring," *IEEE Trans. Biomed. Eng.*, vol. 59, no. 5, pp. 1472–1479, May 2012.

[22] T. Kim, J. Park, J. Sohn, D. Cho, and S. Jeon, "Bioinspired, highly stretchable, and conductive dry adhesives based on 1D–2D hybrid carbon nanocomposites for all-in-one ECG electrodes," *ACS Nano*, vol. 10, no. 4, pp. 4770–4778, 2016.

[23] I. Park *et al.*, "Nanoscale patterning and electronics on flexible substrate by direct nanoimprinting of metallic nanoparticles," *Adv. Mater.*, vol. 20, no. 3, pp. 489–496, 2008.

[24] B. Y. Ahn *et al.*, "Omnidirectional printing of flexible, stretchable, and spanning silver microelectrodes," *Science*, vol. 323, no. 5921, pp. 1590–1593, 2009.

[25] K. Oh, B. H. Byoung, J. K. Hwang, H. Lee, S. Lm, and M. M. Sung, "Water-mediated al metal transfer printing with contact inking for fabrication of thin-film transistors," *Small*, vol. 5, no. 5, pp. 558–561, Mar. 2009.

[26] J. W. Boley, E. L. White, G. T.-C. Chiu, and R. K. Kramer, "Direct writing of gallium-indium alloy for stretchable electronics," *Adv. Funct. Mater.*, vol. 24, no. 23, pp. 3501–3507, Feb. 2014.

[27] E. Forvi *et al.*, "Preliminary technological assessment of microneedles-based dry electrodes for biopotential monitoring in clinical examinations," *Sens. Actuators A, Phys.*, vol. 180, pp. 177–186, Jun. 2012.

[28] J. Kim and W. S. Kim, "A paired stretchable printed sensor system for ambulatory blood pressure monitoring," *Sens. Actuators A, Phys.*, vol. 238, no. 1, pp. 329–336, Feb. 2016.

[29] J. Kim, K. Wubs, B.-S. Bae, and W. S. Kim, "Direct stamping of silver nanoparticles toward residue-free thick electrode," *Sci. Technol. Adv. Mater.*, vol. 13, no. 3, p. 035004, Jun. 2012.

[30] W. C. Bae, D. Kim, M. K. Kwak, L. Ha, S. M. Kang, and K. Y. Sun, "Enhanced skin adhesive patch with modulus-tunable composite micropillars," *Adv. Healthcare Mater.*, vol. 2, no. 1, pp. 109–113, Jan. 2013.

[31] D. Y. Lee, D. H. Lee, S. G. Lee, and K. Cho, "Hierarchical gecko-inspired nanohairs with a high aspect ratio induced by nanoyielding," *Soft Matter*, vol. 8, pp. 4905–4910, Feb. 2012.

[32] H.-L. Peng, J.-Q. Liu, Y.-Z. Dong, B. Yang, X. Chen, and C.-S. Yang, "Parylene-based flexible dry electrode for bioptential recording," *Sens. Actuators B, Chem.*, vol. 231, pp. 1–11, Aug. 2016.

[33] H.-L. Peng *et al.*, "Flexible dry electrode based on carbon nanotube/polymer hybrid micropillars for biopotential recording," *Sens. Actuators A, Phys.*, vol. 235, no. 1, pp. 48–56, Nov. 2015.

[34] D. Panil, A. Dessi, J. F. Saenz-Cogollo, G. Barabino, B. Fraboni, and A. Bonfiglio, "Fully textile, PEDOT: PSS based electrodes for wearable ECG monitoring systems," *IEEE Trans. Biomed. Eng.*, vol. 63, no. 3, pp. 540–549, Mar. 2016.

[35] X. Guo *et al.*, "A self-wetting paper electrode for ubiquitous bio-potential monitoring," *IEEE Sensors J.*, vol. 17, no. 9, pp. 2654–2661, May 2017.

[36] D. Y. Liu, G. X. Sui, and D. Bhattacharyya, "Synthesis and characterisation of nanocellulose-based polyaniline conducting films," *Compos. Sci. Technol.*, vol. 99, no. 30, pp. 31–36, Jul. 2014.

[37] K. Chiller, B. A. Selkin, and G. J. Murakawa, "Skin microflora and bacterial infections of the skin," *J. Invest. Dermatol. Symp. Proc.*, vol. 6, no. 3, pp. 170–174, Dec. 2001.

[38] L. Yue *et al.*, "Sulfonated bacterial cellulose/polyaniline composite membrane for use as gel polymer electrolyte," *Compos. Sci. Technol.*, vol. 145, no. 16, pp. 122–131, Jun. 2017.

[39] I. Kohsari, Z. Shariatinia, and S. M. Pourmortazavi, "Antibacterial electrospun chitosan–polyethylene oxide nanocomposite mats containing bioactive silver nanoparticles," *Carbohydrate Polym.*, vol. 140, no. 20, pp. 287–298, Apr. 2016.

[40] M. K. Delano and C. G. Sodini, "A long-term wearable electrocardiogram measurement system," in *Proc. IEEE Int. Conf. Body Sensor Netw.*, May 2013, pp. 1–6.

[41] B. A. Reyes *et al.*, "Novel electrodes for underwater ECG monitoring," *IEEE Trans. Biomed. Eng.*, vol. 61, no. 6, pp. 1863–1876, Jun. 2014.

[42] J. Pan and W. J. Tompkins, "A real-time QRS detection algorithm," *IEEE Trans. Biomed. Eng.*, vol. BME-32, no. 3, pp. 230–236, Mar. 1985.
[43] N. Zhang *et al.*, "A novel technique for fetal ECG extraction using single-channel abdominal recording," *Sensors*, vol. 17, no. 3, p. 457, 2017.
[44] H. Wang, E. Zhu, J. Yang, P. Zhou, D. Sun, and W. Tang, "Bacterial cellulose nanofiber-supported polyaniline nanocomposites with flake-shaped morphology as supercapacitor electrodes," *J. Phys. Chem. C.*, vol. 116, no. 24, pp. 13013–13019, 2012.
[45] H. Ding *et al.*, "Biologically derived soft conducting hydrogels using heparin-doped polymer networks," *ACS. Nano*, vol. 8, no. 5, pp. 4348–4357, 2014.
[46] L. Yue *et al.*, "Preparation of a carboxymethylated bacterial cellulose/polyaniline composite gel membrane and its characterization," *RSC Adv.*, vol. 6., no. 73, pp. 68599–68605, Mar. 2016.
[47] S. Kaitainen, A. Kutvonen, M. Suvanto, T. T. Pakkanen, R. Lappalainen, and S. Myllymaa, "Liquid silicone rubber (LSR)-based dry bioelectrodes: The effect of surface micropillar structuring and silver coating on contact impedance," *Sens. Actuators A, Phys.*, vol. 206, pp. 22–29, Feb. 2014.
[48] A. Zucca *et al.*, "Tattoo conductive polymer nanosheets for skin-contact applications," *Adv. Healthcare Mater.*, vol. 4, no. 7, pp. 983–990, May 2015.
[49] Y. Meng, Z. B. Li, X. Chen, and J. P. Chen, "A flexible dry micro-dome electrode for ECG monitoring," *Microsyst. Technol.*, vol. 21, no. 6, pp. 1241–1248, 2015.
[50] K. Lin, X. Wang, X. Zhang, B. Wang, J. Huang, and F. Huang, "An FPC based flexible dry electrode with stacked double-micro-domes array for wearable biopotential recording system," *Microsyst. Technol.*, vol. 23, no. 5, pp. 1443–1451, May 2017.
[51] J.-Y. Baek, J.-H. An, J.-M. Choi, K.-S. Park, and S.-H. Lee, "Flexible polymeric dry electrodes for the long-term monitoring of ECG," *Sens. Actuators A, Phys.*, vol. 143, no. 2, pp. 423–429, 2008.
[52] Z. Shi, G. O. Phillips, and G. Yang, "Nanocellulose electroconductive composites," *Nanoscale*, vol. 5, pp. 3194–3201, Feb. 2013.
[53] X. Chen, F. Yuan, H. Zhang, Y. Huang, J. Yang, and D. Sun, "Recent approaches and future prospects of bacterial cellulose-based electroconductive materials," *J. Mater. Sci.*, vol. 51, no. 12, pp. 5573–5588, Jun. 2016.