

## Accepted Manuscript

Title: A potentiometric disposable sensor strip for measuring pH in saliva

Author: Claudio Zuliani Giusy Matzeu Dermot Diamond

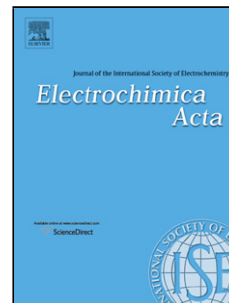
PII: S0013-4686(14)00653-7  
DOI: <http://dx.doi.org/doi:10.1016/j.electacta.2014.03.140>  
Reference: EA 22456

To appear in: *Electrochimica Acta*

Received date: 5-11-2013  
Revised date: 31-1-2014  
Accepted date: 24-3-2014

Please cite this article as: C. Zuliani, G. Matzeu, D. Diamond, A potentiometric disposable sensor strip for measuring pH in saliva, *Electrochimica Acta* (2014), <http://dx.doi.org/10.1016/j.electacta.2014.03.140>

This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.



# A potentiometric disposable sensor strip for measuring pH in saliva

Claudio Zuliani\*<sup>#</sup>, Giusy Matzeu<sup>#</sup>, Dermot Diamond\*

*Centre for Sensor Web Technologies, Dublin City University, Glasnevin, Dublin 9, Ireland*

<sup>#</sup>authors contributed equally to this work

Tel: +353 1 700 6404

Email: [c.zuliani@imperial.ac.uk](mailto:c.zuliani@imperial.ac.uk); [dermot.diamond@dcu.ie](mailto:dermot.diamond@dcu.ie)

## **Abstract.**

In this paper, the preparation of a potentiometric strip for pH monitoring in saliva samples is reported. The potentiometric strip consists of a solid contact pH-selective and of a solid-contact ionogel reference electrode prepared on a dual screen printed substrate. The screen printing protocols, *i.e.*, type of inks and number of deposits, were adjusted to relatively improve the batch reproducibility and the stability of the pH sensor. The pH of real saliva samples was monitored using the optimised potentiometric strip, and results were validated through parallel measurements with a standard laboratory method.

**Keywords:** pH sensor, reference electrode, ionic liquid, saliva, screen printing

## 1. Introduction

Ion-selective electrodes (ISEs) have been widely applied to detect many ions in blood [1-3] and saliva [4-8] samples. The ISEs may have a great potential for a wide range of analytical applications, particularly when produced in a solid-state form since this can drive down the unit cost. This peculiarity could also enable the sensors to be produced in a variety of form factors which in turn can open new application concepts, such as wearable sensors for real-time monitoring of physiological fluids like sweat or saliva [9]. Solid-contact ion-selective electrodes (solid-contact-ISEs) have attracted great attention since the internal reference electrode and the inner filling solution are replaced by a solid material [10]. However, in spite of extensive research in the area, it seems that obtaining solid-contact-ISEs with reproducible standard potentials is still a significant challenge [10] and much research is still focused on the use of standard macroelectrodes rather than low-cost solid-state electrodes.

In order to accomplish the potentiometric measurement, a reference electrode compatible with mass production, *e.g.*, planar-type design, screen printing technologies, solid state functional components, is required. The most challenging applications are those that require continuous use over longer periods of time [9, 11, 12]. For instance, Rius-Ruiz et al. [12] developed a potentiometric strip to monitor the potassium content in samples of saliva and soft drinks diluted with buffers. Accessible body fluids such as saliva represent an interesting medium for the realization of body-sensing, as these fluids also contain important personal physiological information [13-20]. Continuous monitoring of pH in saliva would be extremely beneficial for maintaining healthy mouth conditions, *e.g.*, degradation of tooth enamel [21] and detection of Gastroesophageal Reflux Disease (GERD) [22-24], as well as drug activity, as in some cases this can depend on saliva pH [17].

In this manuscript, we report on the preparation of a potentiometric strip based on a planar screen printed substrate for the direct measurement of pH in saliva samples. The strip contains two electrodes, one sensitive to pH and the other one operating as a reference electrode. pH measurements of saliva samples were validated using a commercial glass electrode.

## 2. Experimental

### 2.1 Materials

Silver ink and C10903P14 as carbon ink obtained from Gwent Electronic Materials (Pontypool, UK) and Acheson Electrodag 452 SS from Henkel (Irvine, CA, USA) as dielectric were used for the preparation of screen printed electrodes. C2030519P4 and D50706D2 from Gwent Electronic Materials were used respectively as carbon and dielectric inks in the preparation of the silver-free screen printed electrodes. 175  $\mu\text{m}$  thick PET sheets were obtained from HiFi (Dublin, Ireland) or from MacDermid (Oxon, UK) and they were used without further treatment. Potassium and sodium chloride, 3,4-ethylenedioxythiophene (EDOT, 97%), poly(3-octylthiophene-2,5-diyl) regiorandum (POT), high molecular weight poly(vinyl chloride) (PVC), bis(2-ethylhexyl) sebacate (DOS,  $\geq 97\%$ ), tetrahydrofuran (THF,  $\geq 99.5\%$ ), 2-nitrophenyl octyl ether (o-NPOE,  $> 99\%$ ), 4-nonadecylpyridine selectophore (hydrogen ionophore II), potassium tetrakis(4-chlorophenyl)borate (KTfPB), citric acid, sodium phosphate dibasic, boric acid, nitric acid and standard buffers (pH 4, 5, 6, 7, 8), 2,2-dimethoxy-2-phenylacetophenone (DMPP,  $> 99\%$ ), butyl-acrylate ( $> 99\%$ ) and 1,6-hexanediol diacrylate (HDDA, 80%) were purchased from Sigma-Aldrich (Dublin, Ireland). When possible, they were of selectophore grade or trace metal standard. N-decyl-methacrylate was obtained from Polysciences (Northampton, UK), chloroform ( $> 99\%$ ) and ethanol (EtOH) from Fisher Scientific (Dublin, Ireland). 1-ethyl-3-methylimidazolium [EMIM]<sup>+</sup> bis(trifluoromethylsulfonyl)imide [NTf<sub>2</sub>]<sup>-</sup>, and [EMIM]<sup>+</sup> tris(pentafluoroethyl)trifluorophosphate [FAP]<sup>-</sup> were obtained from VWR (Dublin, Ireland). All chemicals were used as received. 0.8 mm thick adhesive poly foam strips for the preparation of gaskets were from Radionics (Dublin, Ireland). Deionised water with resistivity of 18.2 M $\Omega$  cm was obtained with a Milli-Q reagent-grade water system and it was used for making aqueous solutions. The pH of deionised water was  $\sim 6.5$ , measured with a commercial pH electrode.

### 2.2 Methods and Procedures.

All screen printed electrodes, SPEs, with and without silver tracks, were realized using a DEK 248 printer (Weymouth, UK). Single SPEs consisted of a carbon disk having a geometric area of  $\sim 0.062 \pm 0.002 \text{ cm}^2$ . For the silver-free SPEs, two layers of the D50706D2 dielectric ink were formed at the speed of 10  $\text{cm s}^{-1}$  which represented a good compromise between the quality of the insulation, and time and materials spent for an additional print. The C2030519P4 carbon ink was printed at 30  $\text{cm s}^{-1}$ . The squeegee

“pressure” value was adjusted to 14 and 10 kg, respectively, for the printing of D50706D2 and C2030519P4, and table/screen separation was the closest achievable with the instrument, i.e., ~ 1-2 mm. Each printed layer was annealed according to the supplier specifications, i.e., silver ink: 5 minutes at 120°C, C10903P14: 15 minutes at 120°C, C2030519P4: 12 minutes at 80°C, D50706D2: 30 minutes at 80°C. In case of multiple prints with same ink, curing was repeated in between printing of individual layers. Silver-free dual electrodes were printed according to the procedure followed for the single silver-free screen printed substrates with the main differences being the design pattern and the individual geometrical area, i.e.,  $\sim 0.045 \pm 0.002 \text{ cm}^2$ .

The pH sensitive membrane was prepared as previously reported [25], drop casting a total amount of 24.5  $\mu\text{L}$  of capping membrane according to the following sequence; 1  $\mu\text{L}$ , 1.5  $\mu\text{L}$ , 8 additions of 2  $\mu\text{L}$ , 2 additions of 3  $\mu\text{L}$ . In order to improve reproducibility, a new design was developed. The pH-ISEs, PMMA (125 or 500  $\mu\text{m}$  thick) and PSA films (50  $\mu\text{m}$  thick) were cut by means of a CO<sub>2</sub> Laser Micro-machining Light Deck (Optec, Belgium) and laminated onto the SPEs by a thermal roller laminator Titan-110 (GBC Films, USA). By combining the PSA film with the 125  $\mu\text{m}$  and 500  $\mu\text{m}$  PMMA film, two gaskets with a 2.4 mm diameter and a nominal depth of 175 and 550  $\mu\text{m}$  were prepared. 5.25  $\mu\text{L}$  of a 2.75 mg mL<sup>-1</sup> solution of POT in chloroform (stirred overnight) were drop cast on top of the carbon layer within the gasket. With the thinner gasket, a total of 8  $\mu\text{L}$  of membrane cocktail solution was drop cast (two additions of 1  $\mu\text{L}$  and 3 additions of 2  $\mu\text{L}$ ), while for the thicker gasket the total amount was 22  $\mu\text{L}$  (1  $\mu\text{L}$  once, 9 additions of 2  $\mu\text{L}$  and 3  $\mu\text{L}$ , once).

The ISEs were conditioned overnight in a solution that was 1 mM each in boric acid, citric acid and sodium phosphate dibasic. The buffer solutions used for the pH sensors calibration were standard buffers (pH 4, 5, 6, 7, 8) or they were made with an opportune amount of citric acid and disodium phosphate, in order to adjust the pH to the desired values during the simulation of saliva. In the last case, the buffer solutions contained 10 mM NaCl and 20 mM KCl as a constant electrolyte background to mimic saliva composition. The solid-contact ionogel reference electrodes (solid-contact-REs) were prepared as reported elsewhere [26] using the silver-free screen printed substrates. In summary, the solid contact was a PEDOT layer electrochemically grown on the carbon electrodes from a 0.05 M EDOT solution in [EMIM]<sup>+</sup>[FAP]<sup>-</sup> while the membrane was obtained by entrapping [HMIM]<sup>+</sup>[FAP]<sup>-</sup> in a poly(butyl-co-decylmethacrylate) phase.

### 2.3 Instrumentation and Software.

The potentiometric measurements were recorded using the EMF-16 voltmeter from Lawson Labs, USA. In-house made silver/silver chloride (saturated KCl) or calomel (IJcambria, UK) electrodes were used as references. A CHI600 or 760D potentiostat (CH Instruments, USA) was used to electro-deposit PEDOT. A pH meter (SympHony SP70P) from VWR was used to validate pH measurements of sublingual saliva samples which were collected using a sterile pipette from volunteers. Photo-polymerization of the acrylate monomers was obtained using the CL-1000 ultraviolet cross-linker UVP. Thickness and profilometer measurements were recorded using a Dektak 150 Profilometer from Veeco (USA). All the Scanning Electron Microscopy (SEM) images were captured with the Hitachi S3000N in the secondary electron mode.

### 3. Results and Discussion

pH sensors were initially prepared using screen printed substrates in which a layer of silver ink was deposited between a PET substrate and a carbon upper layer. Ion-selective electrodes with a solid-contact formed by PEDOT electrodeposited from aqueous KCl (Figure S1), [EMIM]<sup>+</sup>[NTf<sub>2</sub>]<sup>-</sup> or [EMIM]<sup>+</sup>[FAP]<sup>-</sup> (not shown) gave very irreproducible results. POT was preferred as the solid-contact layer for further studies because of the superior reproducibility of the resulting pH sensors. Table 1 presents the calibration slopes and offsets obtained with the pH ISEs fabricated using several types of screen-printed substrates. For all of the sensors in this table, POT was used as solid-contact-layer. pH ISEs prepared from screen printed substrates containing a silver layer showed a sub-Nernstian slope (slope is  $51.9 \pm 0.4$  mV,  $n=4$ ), over the range pH 3-8 when calibrated initially (day 0), see Figure 1a. Their response deteriorated when the electrodes were left in the conditioning solution for 3 days, see Figure 1b. A decrease in linearity was observed ( $R^2$  decreases to  $\leq 0.96$ ) and also the reproducibility within this batch worsened as suggested by the larger standard errors for the average values of slope and offset. Under alkaline conditions, a loss of selectivity of the ion-sensitive membrane is apparent, with the sensor response becoming increasingly governed by other ions in the solutions in accordance with the Nicolsky-Eisenman equation.

It is perhaps important to note that the silver track changed its colour to a darker hue (Figure S2) when sensors were stored in buffer solution for 3 days which may also indicate that some oxidation may be happening [27]. To avoid any concern, pH-sensors were prepared

from screen-printed substrates fabricated using a heat-curable dielectric and without silver ink. Regarding the preparation of carbon screen-printed electrodes, it should be noted that the height profile of carbon layers formed by 1 carbon print was more uniform, i.e., average roughness than those formed by 2 carbon prints, average roughness  $\pm 5$  and  $\pm 7$   $\mu\text{m}$ , respectively, (see Figure S3). In addition, the single print film was thinner with an average thickness of  $11.3 \pm 0.5$   $\mu\text{m}$  compared to  $21.5 \pm 1.8$   $\mu\text{m}$  for the double print carbon layer. In both cases SEM images of the carbon prints were similar to Figure S4.

The use of a gasket was found to be beneficial in terms of confining the drop-cast area, both for the capping membrane and POT deposition, thus generating layers with more reproducible thickness and reproducibility of the sensor response characteristics. Gaskets of two heights were tested in order to allow drop-casting of different amounts of pH sensitive capping PVC membrane, i.e. 8  $\mu\text{L}$  (G8) and 22  $\mu\text{L}$  (G22). The use of G8 gaskets tended to give irreproducible results, which are not discussed further. Table 1 shows that sensors prepared using the G22 gasket, one carbon layer print and the heat-curable dielectric afford good batch reproducibility which persists after 5 days storage in the conditioning solution or after 14 days dry storage with little change of the calibration characteristics. Thus, the pH-sensors in the following section were prepared using this optimised configuration.

The pH-ISEs were combined with disposable ionogel-based liquid-junction-free reference electrodes. These electrodes do not exhibit appreciable changes in potential over the pH range 4-7, and are quite insensitive to potassium and sodium. In fact, when the concentrations of  $\text{K}^+$  and  $\text{Na}^+$  change over two orders of magnitude, i.e., from 1 mM to 0.1 M, the potential of these solid-contact reference electrodes increases by ca. 4-6 mV and 10-15 mV, respectively, against a commercial double-junction reference electrode [26]. In saliva, sodium and potassium levels significantly depend on the area and sampling method, but their concentrations range are always within 1-98 mM and 10-80 mM, respectively [28]. Since the average sodium and potassium concentrations in whole unstimulated saliva are typically  $8 \pm 3$  mM and  $21 \pm 4$  mM, respectively, the solutions for the pH ISEs calibration were made with similar content of  $\text{Na}^+$  and  $\text{K}^+$ , in order to mimic the saliva composition.

Figure 2 shows a typical calibration curve obtained with the pH and solid-contact ionogel reference electrode (SCI-REs) combined in a potentiometric strip. The device response was linear over the pH range required for saliva analysis; i.e. ca. 5.0 to 7.5. The values of potential obtained with the same strip for 4 saliva samples are also plotted. Table 2 reports other tests done on multiple saliva samples with a batch of 4 potentiometric strips, and the

corresponding values found by the pH-meter. The average relative differences between the values recorded using the potentiometric strips and a pH-meter was  $0.2 \pm 0.2$  ( $n=13$ ). It is important to note that the signal of the potentiometric strip placed in contact with the saliva sample became stable after 2 minutes showing an average drift of  $-0.34 \text{ mV/min} \pm 0.08$  (standard error,  $n=17$ ) over the remaining measuring time, i.e., 10-15 minutes. Figure 2 also demonstrates that once the saliva was in contact with the potentiometric strip, both the pH and the SCI-RE electrodes were affected to a similar extent. In fact, this figure compares the values of potentials obtained with the  $\text{H}^+$ -ISE against the solid-contact ionogel reference electrode and against an Ag/AgCl electrode during the measurements in the calibration solutions and saliva samples. There was no significant difference in the calibration slopes, as expected, and the difference in the offset values depended on the bias between the two reference electrodes. More significantly, it is apparent that the accuracy in measuring the pH in saliva samples was greater when the potentials of the pH electrodes were measured against the SCI-RE rather than the Ag/AgCl RE. This occurred because the potential of both of the electrodes in the potentiometric strip decreased to a similar extent during the first minute of immersion, and therefore self-compensated the potential drop. A possible explanation of the above behaviour is that the adsorption of saliva components on the two polymer membranes is similar, while it differs for the Vycor® tip employed for the Ag/AgCl electrode.

A common limitation of solid-contact-ISEs is the need to re-condition the electrodes after a period of dry-storage. Although this period cannot be universally defined and it is often omitted or not reported in the literature [29], commercial solid-contact-ISEs recommend re-conditioning of at least 1 hour before commencing the measurement [30]. Indeed, Table 2 shows the pH values of saliva samples obtained with two potentiometric strips which were re-conditioned for 40 minutes in a buffer solution after a 5 hour dry-storage period. The average percentage difference between the pH values measured with the glass electrode and the ones extrapolated from the 1-point calibration of the above strips was  $0.2 \pm 0.1$  ( $n=5$ )

It is noteworthy to highlight that in Table 2, some saliva samples were collected before and after consumption of 150 mL of a soft drink and that these samples did not show significant changes in pH. This result probably depends on the buffer ability of saliva [28]. Thus, in order to demonstrate the ability of the potentiometric strips to monitor an extended pH window in real saliva, the pH of a sample was monitored while titrating it with a solution of 0.1 M HCl. Figure 3 shows the trace of the potentials recorded with the  $\text{H}^+$ -ISE, the SCI-RE and the potentiometric strip. In this figure the pH spontaneously increased from 7.02 to 7.06



in the first 10 minutes as measured by means of a pH glass electrode. Interestingly, this pH increase agrees with the slight decrease of the potential of the potentiometric strip. Significantly, the stability of the disposable reference electrode of the potentiometric strip was good since its maximum variation during the titration experiment was  $\pm 6$  mV. The spiking of HCl solution was performed as indicated by the arrows and the pH of saliva was measured with the pH-meter 2 minutes after this addition. Table 3 compares these values with the ones extrapolated from one-point calibration of the potentiometric strip. Indeed, in the latter case the device offset was determined by reading the potential of the strip in contact with a standard solution at pH=7.5, and taking the average slope obtained with the sensors batch reported in Table 2 as device sensitivity. Table 3 demonstrates that the largest bias between the two methods in evaluating the saliva pH in the range 4-7 is smaller than 0.5 units. This small error gives the opportunity for integrating the potentiometric strip into wearable platforms and future work will address this implementation.

## Conclusions

This work highlights the preparation of a disposable potentiometric strip which can monitor pH in sublingual saliva samples in good correlation with data from a commercial pH meter. It was also demonstrated that the potentiometric strip withstands a short-period of dry-storage and that, after a short re-conditioning step, pH measurements of saliva samples obtained employing 1-point calibration are reliable. The small differences in the saliva pH readings given by the pH-meter and the potentiometric strips (with both the calibration methods, i.e., 1- or 5-points) are acceptable for applications targeting the monitoring of in-mouth pH, since the analytical method has to discriminate among saliva samples having wide pH regions: acid (pH 5.0-5.8), moderately acidic (pH 6.0-6.6) and healthy saliva (pH 6.8-7.8). Future work will focus on the integration of the strip into a suitable in-mouth wearable device.

## Acknowledgments

Science Foundation Ireland is gratefully acknowledged for supporting this project under the grants 07/CE/I1147 and 03/IN.3/1361/EC07. Authors would like also to thank Dr. Fiachra Collins for the CAD drawings of the screen patterns.

**FIG CAPTIONS**

**Figure 1.** (a) 1<sup>st</sup> day and (b) 3<sup>rd</sup> day calibration curves of four pH sensors based on SPE containing a silver layer as contact pad. Electrodes were left in the conditioning solution in between measurements. Data points labels: (●) series 1, (■) series 2, (◆) series 3, and (▲) series 4. Linear fit labels: (—) series 1, (– –) series 2, (· · ·) series 3, and (– · · –) series 4. In (b) the linear fit for the (▲) series 4 is not reported since the low  $R^2$  value which was equal to 0.75.

**Table 1.** Calibration data of pH ISEs prepared using different screen printing protocols. The average slope and offset are reported together with their standard error and % change. Number of sensors for each batch is also specified. Day 0 refers to the electrodes being calibrated after the standard conditioning step. In between the re-calibrations sensors were stored in the conditioning solution for the number of days indicated in the table, except in the case labelled (\*) in which the electrodes were kept dry for an additional 14 days before the measurement.

**Figure 2.** Calibration curve for a pH electrode obtained by plotting its potential bias against (– · –) the solid-contact-RE or (—) against the Ag/AgCl electrode. The potential values obtained with the pH electrode in contact with the calibration solutions and with the saliva samples are reported against the solid-contact-RE (○) and (●), respectively, or against the Ag/AgCl electrode (□) and (■), respectively.

**Table 2.** pH data of saliva samples measured with a commercial glass electrode and the optimized potentiometric strips. At least, 2 saliva samples were tested with each platform (each potentiometric platform is highlighted by a bold box). In case of 1-point calibration, the potentiometric strips were stored dried for 5 hours and then re-conditioned in a buffer at pH=5.7 with a background of 10 mM of NaCl and 20 mM of KCl for 40 minutes, before testing the samples.

**Figure 3.** Traces of electrode potential obtained with the (—) H<sup>+</sup>-ISE and (···) solid-contact-ionogel RE against a Ag/AgCl reference immersed into a saliva sample while adding stepwise a solution of 0.1 M HCl as indicated by the arrows on the graph. The right y-axis refers to the trace of the potential of the potentiometric strip (– –) obtained by plotting H<sup>+</sup>-ISE against SCI-RE.

**Table 3.** pH values of a saliva sample as measured with a glass electrode and a potentiometric strip during its titration with 0.1 M HCl. In the latter case, the values were

obtained by averaging the trace of the potential recorded for 3 minutes after 1.5 minutes from the HCl spiking and using one-point calibration. The one-point calibration was obtained from measuring the device potential once in contact with a solution at pH=7.5 containing 10 mM NaCl and 20 mM KCl and using the average slope of the potentiometric strips reported in the previous table as device sensitivity.

Accepted Manuscript

## FIGURES

FIGURE 1

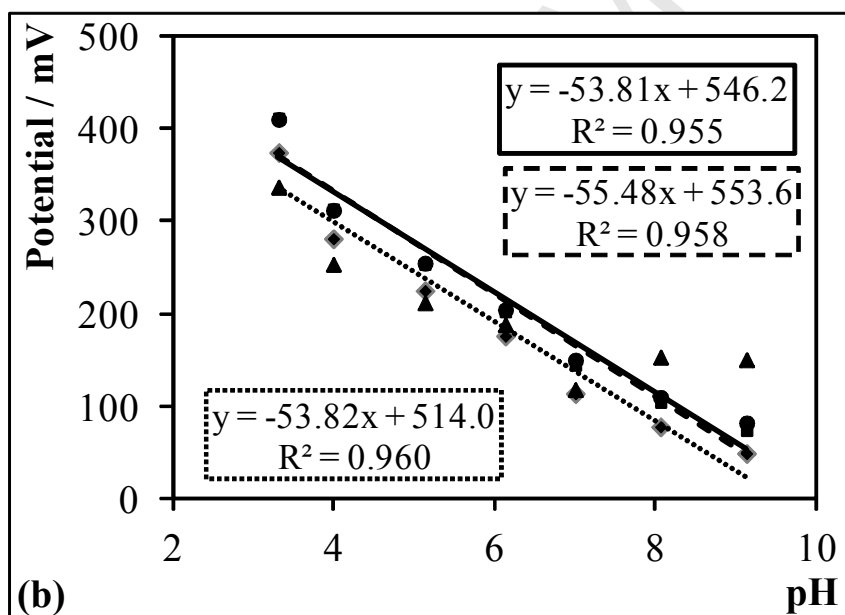
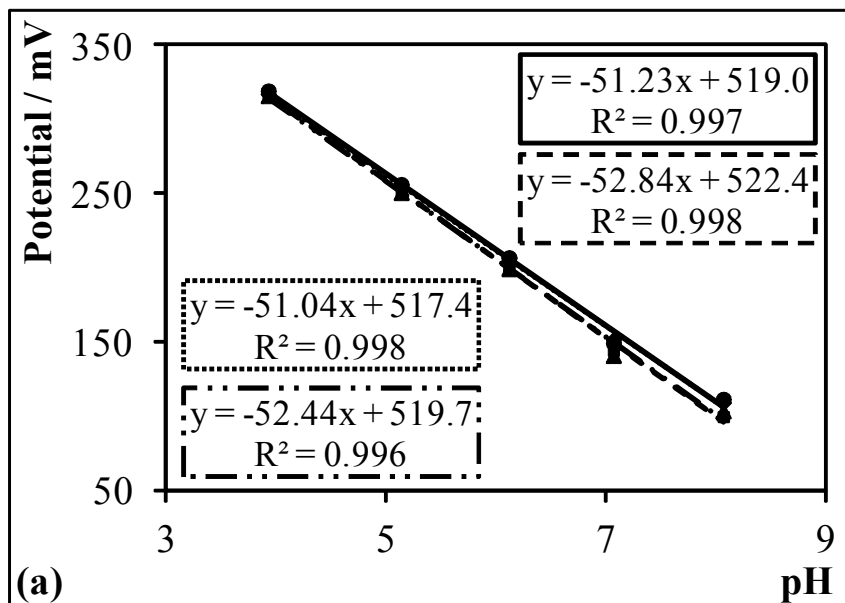


TABLE 1

Configuration	Calibration (day)	Slope (mV/pH)	Offset (mV)	% Slope change	% Offset change
Ag/Carbon/POT	0 (n=4)	-51.9 ± 0.4	519.6 ± 1.0	/	/
Ag/Carbon/POT	4 (n=3)	-54.4 ± 0.6	537.9 ± 12.2	4.8	3.5
Carbon/POT/G22	0 (n=4)	-53.4 ± 0.3	524.0 ± 2.0	/	/
Carbon/POT/G22	6 (n=3)	-55.8 ± 1.1	565.0 ± 7.7	4.5	7.8
Carbon/POT/G22*	19 (n=3)	-55.6 ± 2.4	544.6 ± 10.7	-0.4	-3.6
Carbon/POT/G22	0 (n=4)	-53.5 ± 0.6	537.5 ± 5.3	/	/

FIGURE 2

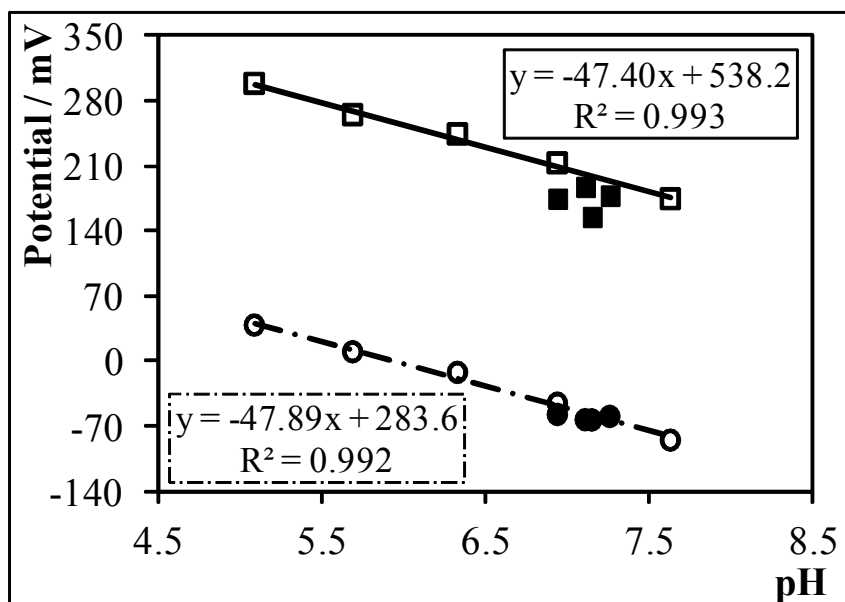


TABLE 2

5-point calibration			1-point calibration		
pH meter	pH sensor	$\Delta$ pH	pH meter	pH sensor	$\Delta$ pH
7.1	7.3	-0.2	7.0	7.3	-0.3
7.3	7.2	0.1	7.1	7.4	-0.3
7.2	7.3	-0.1	6.9	6.8	0.1
6.9	7.1	-0.2	7.1	7.2	-0.1
7.1	7.3	-0.2	6.9	6.9	0.0
7.2	7.7	-0.5			
6.9	7.4	-0.5			
7.3	7.8	-0.5			

FIGURE 3

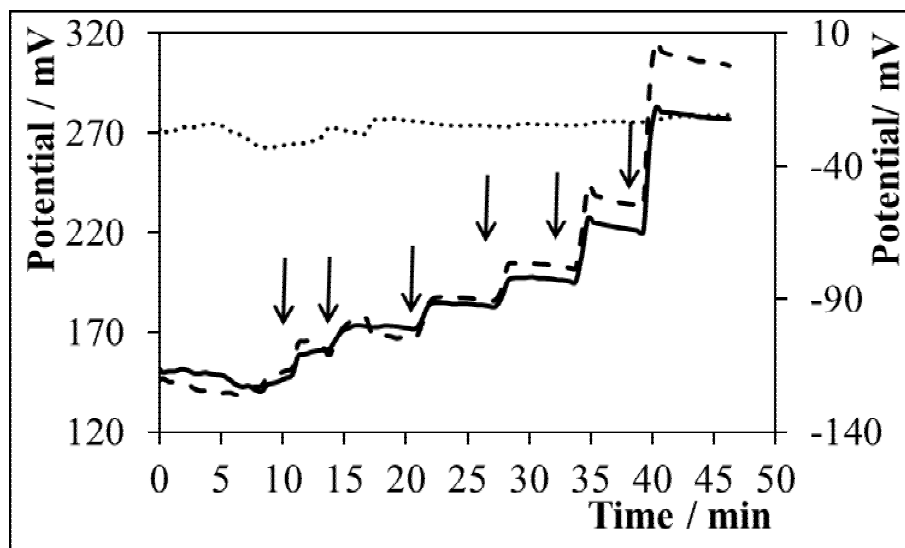




TABLE 3

<b>t/min</b>	<b>pH-meter</b>	<b>pH-Potentiometric strip</b>	<b><math>\Delta</math>pH</b>
10.5	6.5	6.1	0.4
14	6.3	5.9	0.4
20.5	6.1	5.6	0.5
27.5	5.8	5.4	0.4
33.5	5.2	4.9	0.3
39.5	4.1	3.8	0.3

Accepted Manuscript

- [5] M. Khairy, R.O. Kadara, D.K. Kampouris, C.E. Banks, *Analytical Methods*, 2 (2010) 645-649.
- [6] T. Poursaberi, M. Salavati-Niasari, S. Khodabakhsh, L. Hajiagha-Babaei, M. Shamsipur, M. Yousefi, S. Rouhani, M. Ganjali, *Analytical Letters*, 34 (2001) 2621-2632.
- [7] M. Shamsipur, T. Poursaberi, M. Rezapour, M. Ganjali, M. Mousavi, V. Lippolis, D. Montesu, *Electroanalysis*, 16 (2004) 1336-1342.
- [8] O. Kulapina, V. Baraguzina, N. Skoblikova, *Pharmaceutical Chemistry Journal*, 42 (2008) 481-484.
- [9] N. Sawka, L.M. Burke, E.R. Eichner, R.J. Maughan, S.J. Montain, N.S. Stachenfeld, *Med. Sci. Sports Exercise*, 39 (2007).
- [10] J. Bobacka, A. Ivaska, Ion sensors with conducting polymers as ion-to-electron transducers, in: S. Alegret, A. Merkoçi (Eds.) *Comprehensive Analytical Chemistry*, vol. 49, Elsevier, 2007, pp. 73-86.
- [11] S. Anastasova, A. Radu, G. Matzeu, C. Zuliani, D. Diamond, U. Mattinen, J. Bobacka, *Electrochimica Acta*, 73 (2012) 93-97.
- [12] F.X. Rius-Ruiz, G.A. Crespo, D. Bejarano-Nosas, P. Blondeau, J. Riu, F.X. Rius, *Analytical Chemistry*, 83 (2011) 5783-5788.
- [13] D. Diamond, *Analytical Chemistry*, 76 (2004) 278A-286A.
- [14] E. Papacosta, G.P. Nassis, *Journal of Science and Medicine in Sport*, 14 (2011) 424-434.
- [15] G. Ljungberg, T. Ericson, B. Ekblom, D. Birkhed, *Scandinavian Journal of Medicine and Science in Sports*, 7 (1997) 214-219.
- [16] S.A. Khaustova, M.U. Shkurnikov, A.G. Tonevitsky, *Bulletin of Experimental Biology and Medicine*, 149 (2010) 635-639.
- [17] S. Ghimenti, T. Lomonaco, M. Onor, L. Murgia, A. Paolicchi, R. Fuoco, L. Ruocco, G. Pellegrini, M.G. Trivella, F. Di Francesco, *PLOS One*, 6 (2011) e28182.
- [18] A.E. Herr, A.V. Hatch, W.V. Giannobile, D.J. Throckmorton, H.M. Tran, J.S. Brennan, A.K. Singh, *Proc. Oral-Based Diagnostics*, 2007.
- [19] C. Yang, E. Brooks, Y. Li, P. Denny, C. Ho, F. Qi, W. Shi, L. Wolinsky, B. Wu, D. Wong, C. Montemagno, *Lab on a Chip*, 5 (2005) 1017-1023.
- [20] K. Yoda, K. Shimazaki, Y. Ueda, *Annals of the New York Academy of Sciences*, 864 (1998) 600-604.
- [21] A. Millward, L. Shaw, E. Harrington, A. Smith, *Caries Research*, 31 (1997) 44-49.
- [22] C.A. Eckley, H.O. Costa, *Brazilian journal of otorhinolaryngology*, 72 (2006) 55-60.
- [23] M. Bouchoucha, F. Callais, P. Renard, O. Ekindjian, P. Cugnenc, J. Barbier, *Archives of Physiology and Biochemistry*, 105 (1997) 19-26.
- [24] D. Lazarchik, S. Filler, *American Journal of Medicine*, 103 (1997) 107S-113S.
- [25] C. Fay, S. Anastasova, C. Slater, S.T. Buda, R. Shepherd, B. Corcoran, N.E. O'Connor, G.G. Wallace, A. Radu, D. Diamond, *IEEE Sensors Journal* 11 (2011) 2374-2382.
- [26] C. Zuliani, G. Matzeu, D. Diamond, Submitted to *Talanta* (2013).
- [27] T.J. Smith, K.J. Stevenson, Reference Electrodes, in: C.G. Zoski (Ed.) *Handbook of Electrochemistry*, Elsevier, 2007, pp. 92.
- [28] W.F. Boron, E.L. Boulpaep, Pancreatic and Salivary Glands, in: *Medical Physiology*, Elsevier, 2012, pp. 927-931.
- [29] E. Lindner, R. Gyurcsányi, *Journal of Solid State Electrochemistry*, 13 (2009) 51-68.
- [30] B. Schazmann, D. Morris, C. Slater, S. Beirne, C. Fay, R. Reuveny, N. Moyna, D. Diamond, *Analytical Methods*, 2 (2010) 342-348.