Available online at www.sciencedirect.com

ScienceDirect

Procedia Engineering 5 (2010) 816–819

Procedia Engineering

www.elsevier.com/locate/procedia

Proc. Eurosensors XXIV, September 5-8, 2010, Linz, Austria

An Automatic Offset Correction Platform for High-Throughput Ion-Channel Electrophysiology

F. Thei[a,]*, M. Rossi[a], M. Bennati[a], M. Crescentini[a], and M. Tartagni[a]

[a]*ARCES – Advanced Research Center on Electronic Systems, Second School of Engineering, University of Bologna, Via Venezia 52, Cesena (FC) 47521, Italy*

Abstract

High-throughput ion channel screening for drug discovery is at the base of the recent shift of resources in the pharmaceutical industry towards addressing drug safety issues earlier in the discovery process. Very few examples of parallel ion-channel recording platforms are currently present in literature, due to the complexity of the setup. However, single-junction Ag/AgCl electrodes suffer of intrinsic voltage offsets, due to the electrode-buffer interface variability. This is very critical, since ion-channel recording requires high accuracy (pA resolution) within the full-scale (nA range), limiting the operability of the measurement, especially on a multi-channel approach. This paper presents an automatic offset correction system fully implemented on a lipid bilayer membrane platform. The platform allows offset-free recording of ion-channel signals acquired and displayed by means of a graphical user interface.

Keywords: Ag/AgCl electrodes; Ion-Channel;

1. Introduction

The voltage offset variability of the interface between the Ag/AgCl electrodes and the KCl buffer solution in the biosensor devices, schematically represented in Fig. 1, is a well know electrochemical problem, especially in voltammetry, patch clamp and ion channels acquiring systems [1].

Nomenclature

ΔΣ ADC Delta Sigma Analogic to Digital Converter

BLMs artificial Bilayer Lipid Membranes

* Corresponding author: Federico Thei, Second School of Engineering, University of Bologna, Campus of Cesena, Via Venezia 52 – I-47521 Cesena (IT); Tel.: +39-0547-634747; fax: +39-0547-339208; *e-mail address*: fthei@arces.unibo.it.

1877-7058
doi:10.1016/j.proeng.2010.09.233

Ag/AgCl	Silver/Silver Chloride electrode
KCl	Potassium Chloride buffer solution
LPF	Low Pass Filter

Different approaches have been proposed to solve this problem, such as three electrode systems (Ag/AgCl plus Au or Pt electrode [2]), liquid junction, and agar-salt bridge [3].However, these approaches are usually used into single or multiplexed acquisition systems, where a single measurement instrument is switched between the channels without taking into account crosstalk and routing issues arising on concurrent signal acquisition on high-throughput 2D arrays.

The crosstalk at the amplifier input stage of the ΔΣ ADC is the effect of the array approach, where a single signal feeds all the sensors of the array. Particularly, small differences between sensors, detectable through variations of the equivalent impedance, are mapped into relevant offset variations at the input of the ADCs. Moreover, the input of the ΔΣ ADC presents different impedances, due to the intrinsic tolerances of the chip fabrication process parameters. These differences affect the measured current, especially during the initial steps of the acquiring process, where a zero input current is required to initialize the system. On the other hands, unknown offsets over the supply signal of the biosensor affect the accuracy of the measure, especially on voltage-gated ion-channels.

We present a new systematic approach based on a fully electronic feedback system, employing single-junction Ag/AgCl electrodes, capable to be implemented into platforms for parallel ion-channel recording [4] (Fig. 2, Fig. 4). The platform is implemented and tested on a printed circuit board hosting the array where one side is dedicated to the electronic components and the other to the microfluidic array (bottom and top view of Fig. 2 (b)).

2. Structure

The first offset correction step consists in searching the voltage signal value that sets to zero the corresponding current signal. The obtained value becomes the virtual ground reference for the selected channel of the system. The proposed approach is based on an automatic voltage offset correction, operating independently and simultaneously on each channel of the array. The offset correction is implemented using the digital output of the ΔΣ ADC employed for the acquisition [5]. Since the ΔΣ ADC output is a digital signal proportional to the oversampled analog input, it is possible to extract the original offset value by integrating the single-bit digital output stream during the setup phase by a LPF stage. To store permanently the offset voltage value provided by the LPF, a digital potentiometer is used, connected between two voltage references in the range of typical Ag/AgCl - buffer electrochemical interface values (hundred of mV). The potentiometer resistance variation is driven by the offset between the LPF of the ΔΣ output (the *mean value* of Fig. 2 (a)) and the virtual input ground, corresponding to Vcc/2, that is the voltage value equivalent to a zero measured input current (Fig. 2 (a)). If the LPF output is greater than the virtual ground, corresponding to the case of a positive input current acquired by the ΔΣ ADC, the comparator output (Fig. 2 (a)) gives a negative saturation, corresponding to a zero logic value (signal $U/\overline{D}$ of Fig. 2 (a)), allowing to reduce the offset voltage to decrease the measured current.

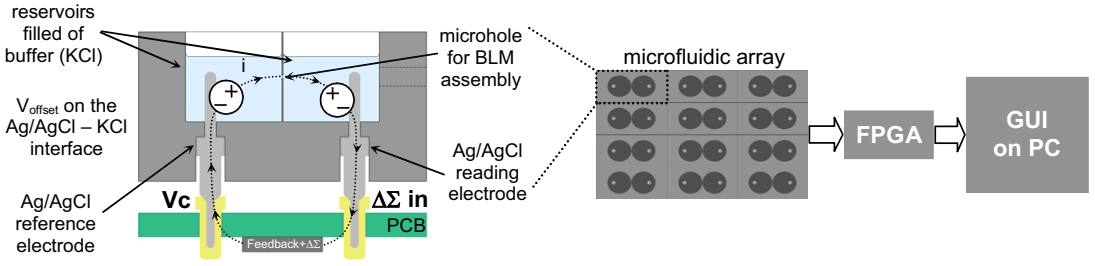

Fig. 1: Details of a single microfluidic block where ion channel signals are recorded, with emphasis on the offset problem and block diagram of the readout system.

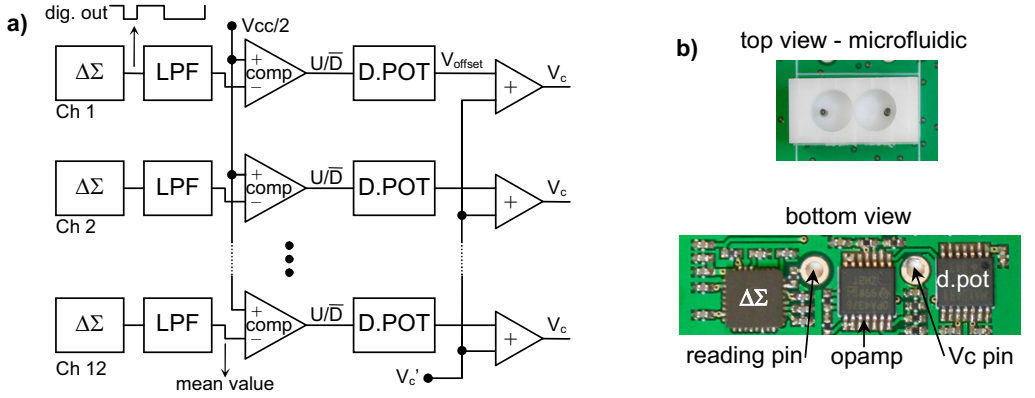

Fig. 2 : (a) The feedback block diagram for Ag/AgCl – buffer interface voltage offset correction within the array approach; (b) detail of one channel section of the PCB implementation with feedback correction and embedded microfluidics.

By this indication, the digital potentiometer implements the reduction of the output voltage (V_{offset} of Fig. 2 (a)), reaching the offset compensation through the negative feedback. The V_{offset} is linearly added to the control voltage (V_c') to get the final Vc, one for each acquired channel. V_c' is the control signal, corresponding to all the channels feeding the sensor array that can be set to 80 mVpp triangular wave @ 10 Hz, used to monitor the BLM formation, or a fixed voltage in the range of ±60 mV, to supply the ion-channel.

3. Test and Measurements

The overall procedure for the automatic feedback correction follows the steps illustrated in Fig. 3. Each of them could be automated: i) the microfluidic chambers are filled with KCl and the pore impedance is electronically verified; ii) offset is corrected and stored into the digital potentiometer; iii) artificial BLM is formed and electronically verified by detection of the impedance modification; iv) ion-channels are inserted into BLMs by means of α-hemolysin proteins; v) signals are recorded and displayed in real time using a PC-based graphical user interface (Fig. 4);

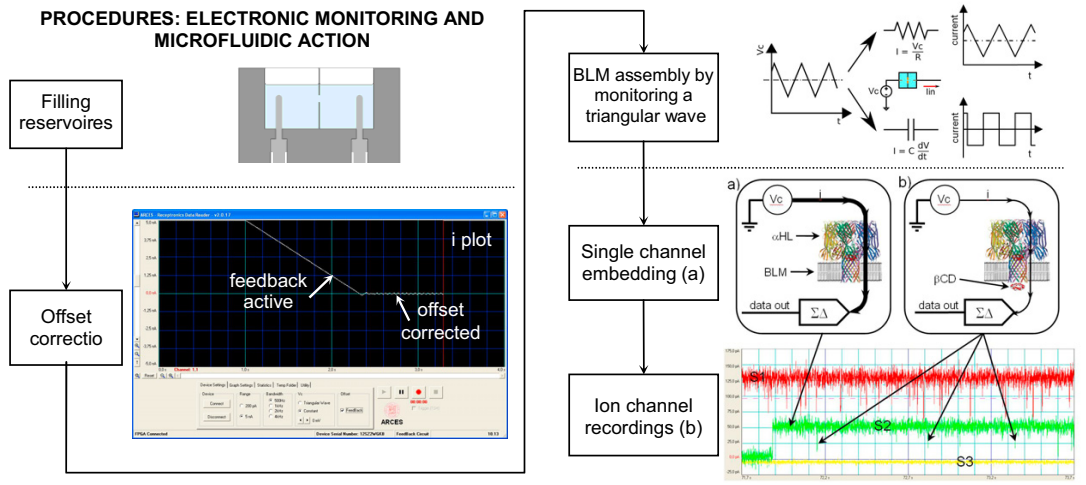

Fig. 3: Flow chart of the overall operational procedures for the automatic offset correction and multiple ion-channel recordings using a lipid bilayer membrane array.

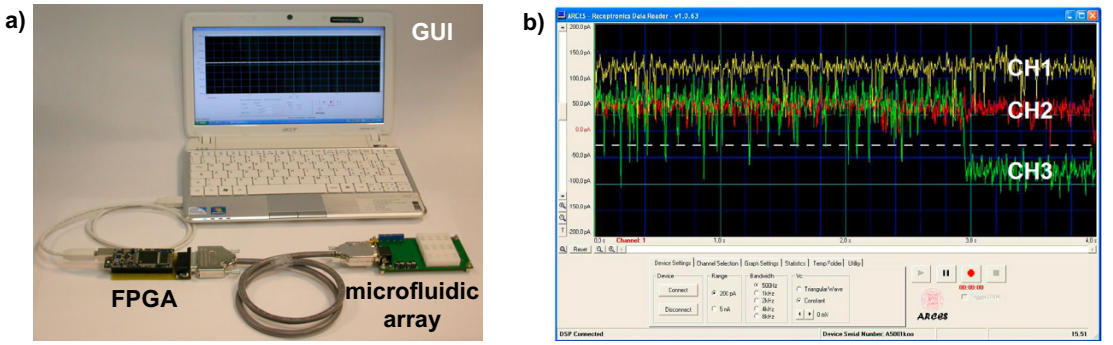

Fig. 4: (a) Overview of the overall platform with graphical user interface on top; (b) Screenshot of the GUI interface displaying the acquisition of three simultaneous single ion-channels in a system without offset compensation. Note CH1, where an offset of 40 mV due to the electrodes is added to the applied control voltage.

The offset setup procedure could be applied on successive phases. The accuracy of the offset correction depends on the resolution, that is number of steps, of the digital potentiometer and from the voltage band gap, centered on the virtual ground potential (Vcc/2), where the digital potentiometer resistance is connected. These two parameters are strictly related to the selected range and resolution of the measured current. For instance, in the range of 5 nA, with a resolution of the ΔΣ ADC of 12 bits, the maximum resolution of the measured current is 1,22 pA. The digital potentiometer used is a MAX 5481, with 1024 steps available. Assuming a voltage band gap of 100 mV, the minimum voltage step to compensate the offset is around 100 μV. Considering that a microfluidic setup present a resistance in the order of 10 MΩ, given by the pore plus the Ag/AgCl electrode resistances, the minimum step in the acquired current is about 10 pA, very close to the zero, considering the 5nA scale

4. Conclusion

An automatic offset correction platform for high-throughput ion-channel electrophysiology is presented. The whole system was implemented, such as the microfluidics, the ADC array inclusive of the feedback circuitry for the automatic offset correction, and the data visualization on a PC. As proof of this concept, an offset correction step was performed during an experimental recording on single-molecule events on parallel single ion-channel signals.

Acknowledgements

This work was supported by the 6th Framework Programme of the European Commission under the contract NMP4-CT-2005-017114 "RECEPTRONICS". We are also grateful to the “Fondazione Cassa dei Risparmi di Forlì” for its support.

References

[1] B. Sakmann and E. Neher “Single Channel Recording” 2nd edition Plenum, New York (1995).

[2] N. Triroj, M. A. Lapierre-Devlin, S. O. Kelley, and R. Beresford “Microfluidic Three-Electrode Cell Array for Low-Current Electrochemical Detection”, IEEE Sensor Journal, Vol. 6, No. 6 (2006), 1395-1402.

[3] X. M. Shao, and J. L. Feldman “Micro-agar salt bridge in patch-clamp electrode holder stabilizes electrode potentials”, Journal of Neuroscience Methods, Vol. 159 (2007), 108–115.

[4] F. Thei, M. Rossi, M. Bennati, M. Crescentini, F. Lodesani, H. Morgan, and M. Tartagni “Parallel Recording of Single Ion Channels: A Heterogeneous System Approach”, IEEE Transactions on Nanotechnology, in-press, 2010.

[5] M. Bennati, F. Thei, M. Rossi, M. Crescentini, G. D’Avino, A. Baschirotto, and M. Tartagni “A Sub-pA Delta-Sigma Current Amplifier for Single Molecule Nanosensors”, ISSCC Digest of Technical Papers, Vol. 52 (2009), 348-350.