Development of an temperature measurement system through impedance to detect changes in single ionic channels using Patch-Clamp technique

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There is a long-standing knowledge that temperature by itself exerts great impact on all biological processes. Temperature gating studies of ion channels by patch clamp recording methods have been made difficult by a lack of methods for rapid alteration and detection of temperature in live cells [1]. Mechanistic study of thermal sensitivity of ion channels requires controlled temperature perturbation. Conventional approaches involve resistive heating or thermal electric heating and cooling [2]. Although currently available techniques represent a significant achievement, the rate of change in temperature may not be sufficient to provide research rapid thermodynamic processes involved in transducing thermosensory.

To address this issue, we developed a closed loop control temperature system with a pulsed infrared diode lasers (as heat generator), a digital ohmmeter to measure temperature changes and a cooling system based on peltier cells (thermoelectric cells) controlled by a pulse width modulator (PWM). These three components were mounted on a container chamber. The aim of this system was to obtain a temperature-controlled pulse and to detect cell responses induced by temperature changes. The digital ohmmeter measures cells impedance (as a temperature indicator) via micropipette electrodes, calculates the initial temperature of the solution is theoretically the initial temperature of the cell using a thermistor automatically selects the gain corresponding to the pipette is used, this gain can be impedance with ranges from 20 MΩ at intervals of 5 MΩ to 5MΩ , taking initial impedance and initial temperature, the system calculates the slope of the line for this pipette and a linear relationship is obtained corresponding to a temperature value, the temperature value is sent serially to the system that controls the laser will serve as the feedback signal, a temperature is selected which is to be heated by means of a digital control system using fuzzy logic and generates a PWM for the hot laser module. The system was designed temperature measurement so that it could be placed on a manipulator and was as close as possible to perform the measurements.

The impedance measurement system has been evaluated through electric simulators and our results corroborate system functionality, getting value correspondence between impedance graphs vs temperature. The cooling system rapidly diminishes the temperature near a cell, does not interfere with patch clamp recordings and does not heat the containment chamber. Our system has the advantage of been a low cost device and to be a useful tool for recording electrical signals from channels of living cells with an adequate temporal resolution through Patch Clamp techniques and under precise temperature control.

[1] Ivan Ditterta, Jan Benedikta, Ladislav Vyklickýa, Katharina Zimmermannb, Peter W. Reehb, Viktorie Vlachováa, "Improved superfusion technique for rapid cooling or heating of cultured cells under patch-clamp conditions" Journal of Neuroscience Methods, Volume 151, Issue 2, 15 March 2006, Pages 178–185.

[2] Jing Yao, Beiying Liu, and Feng Qin, "Rapid Temperature Jump Infrared Diode Laser Irradiation for Patch-Clamp Studies", Biophysical Journal Volume 96 May 2009 3611–3619.