eBook

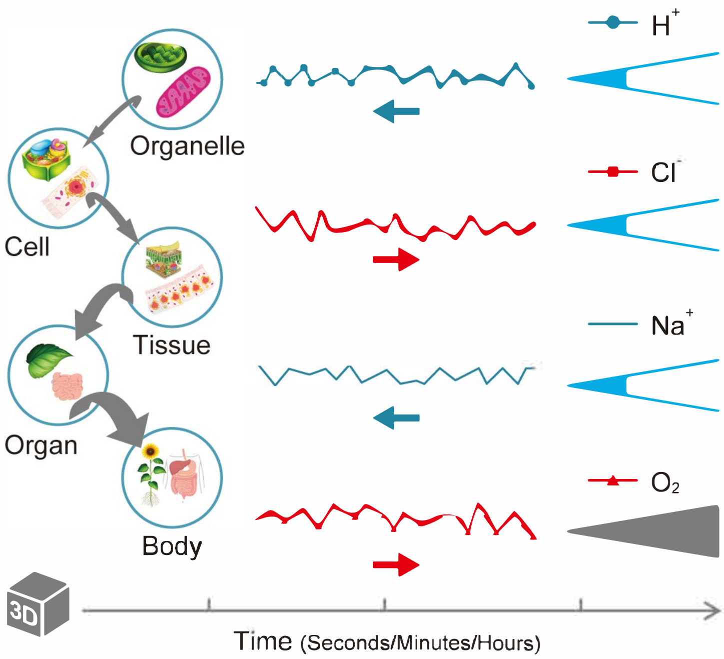
Life is Environment

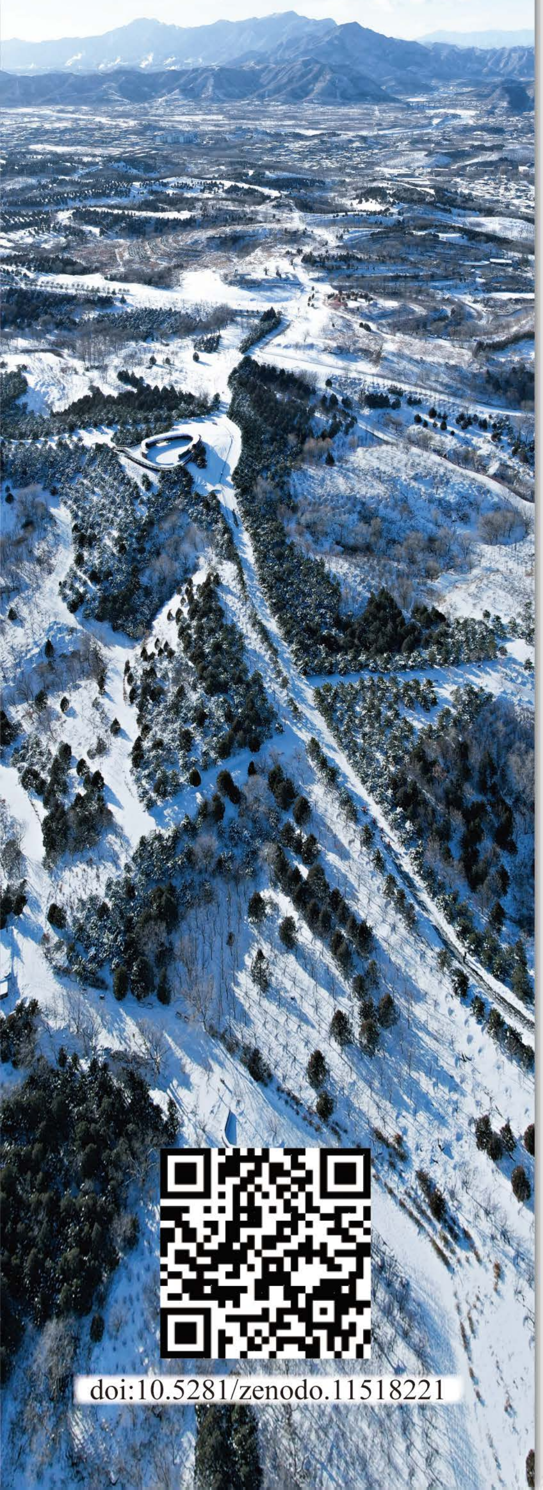
Yue (Jeff) Xu

Founder of YoungerUSA

NMT & Its Applications

Non-invasive Micro-test Technology in Life Sciences





doi:10.5281/zenodo.11518221

2024.5

Ver. 1.0

Acknowledgement

YoungerUSA owes a great debt to so many scientists, professors, students, engineers, business collaborators, competitors and especially our customers all over the world, who have been the inspirations, supports and driving forces behind NMT advancement.

Abstract

Life is an irreversible dynamic equilibrium in which ions and molecules are constantly exchanging between its internal and external environments. NMT: Non-invasive Micro-test Technology can detect this dynamic process by measuring concentration gradients, namely ionic/molecular fluxes, in multi-dimensions in real time and in situ with live samples. NMT has a super sensitivity capable of detecting Ca^{2+} fluxes of a single cell, e.g. a pollen grain, which has been making it an invaluable tool for scientists in many scientific fields. In this life science version, applications from biomedical research to plant and crop sciences, animal science, environmental science, microbiology to marine biology et al. have been covered. Brief NMT history and some new features integrated with current AI capabilities as well as NMT for higher education are also introduced hoping to inspire young scientists to explore this exciting area in the future. Standardization in terms of SOPs stemmed from pioneer NMT scientists, commercialization and industrialization of NMT are also introduced as references for both ambitious bio-entrepreneurs and new people who just came across this technology. The promising imOmics and its related projects, such as Bio-imOmics Project are explained with intention to guide people to fully appreciate the integration capabilities of NMT in the upcoming multi-omics age.

Keywords

NMT, Non-invasive Micro-test Technology, Ions, Molecules, Fluxes, Sensors, Microelectrodes, Transmembrane transport, Microenvironment, Life Sciences, imOmics, BiP, Bio-imOmics Project

Contents

1 Introduction

Ions and molecules are not only the basic particles that make up all things in the world, but also an important medium for information exchange and energy conversion between living and non-living substances and their external environments. From Ion and Molecule Homeostasis (IMH) perspective, it is essential to study ionic/molecular activities in life sciences [1–9]. Diseases of humans, animals, crops, insects and microbes et al. are usually accompanied by abnormal IMHs. Excessive H^+ accumulations, for instance, is the result from lactate formation even in the presence of O_2 in tumor cells demonstrated by Nobel laurate Warburg nearly a century ago[10–14]. It is also well known that a plant will not grow properly if the supply of NH_4^+, NO_3^-, PO_4^{3-}, K^+ among other ions/molecules are out of balance[15–18].

However, it has been a daunting challenge for biologists to fully comprehend the IMHs due to:

a) ionic and molecular information related to IMH have to be collected with live samples in order to make reliable and practical sense of those data;

b) multiple ions/molecules are preferably measured simultaneously in order to construct the relationship network among relevant ion(s)/molecule(s) to a specific physiological process;

c) ionic/molecular data in different levels, i.e. cell, tissue to organ, has to be collected due to the increasing evidence that there are dramatic differences of ionic/molecular transmembrane activities from cellular to tissue and/or organ levels;

d) since biological materials are 3-dimensional per se, 1D or 2D form of data is no longer accurate enough to reflect the biological real 3D world [19–23].

In the last 20+ years, Non-invasive Micro-test Technology (NMT) has been proved to be able to tackle these challenges mentioned above [9,24].

2 NMT: Non-invasive Micro-test Technology

2.1 Principles & features

2.1.1 Two-points measurement scheme

Ionic/molecular fluxes will be detectable when ions/molecules move from higher concentrations to lower concentrations. The concentration differences will then result in ionic/molecular gradients **(Figure 1.)**. In the scenarios of live biological samples, transmembrane activities of ions/molecules will result in ionic/molecular gradients via their effluxes and/or influxes [25].

In order to measure these ions/molecules selectively or specifically in a quantitative way, various sensors made from different materials, such as glass, optical fiber, platinum-iridium alloy, nanoparticles et al., have been developed **(Table 1.)**.

These sensors' fabrications are based on different theories/mechanisms, such as ionophore special characteristics, enzymatic reactions, electrochemistry, fluorescent dyes etc. [26].Due to the special demands of temporal and spatial resolutions, conventional ionic/molecular sensors usually have to be modified to meet the requirements from NMT hardware configuration. Therefore, NMT ionic/molecular micro-sensors are designated to differentiate themselves from those traditional micro-sensors.

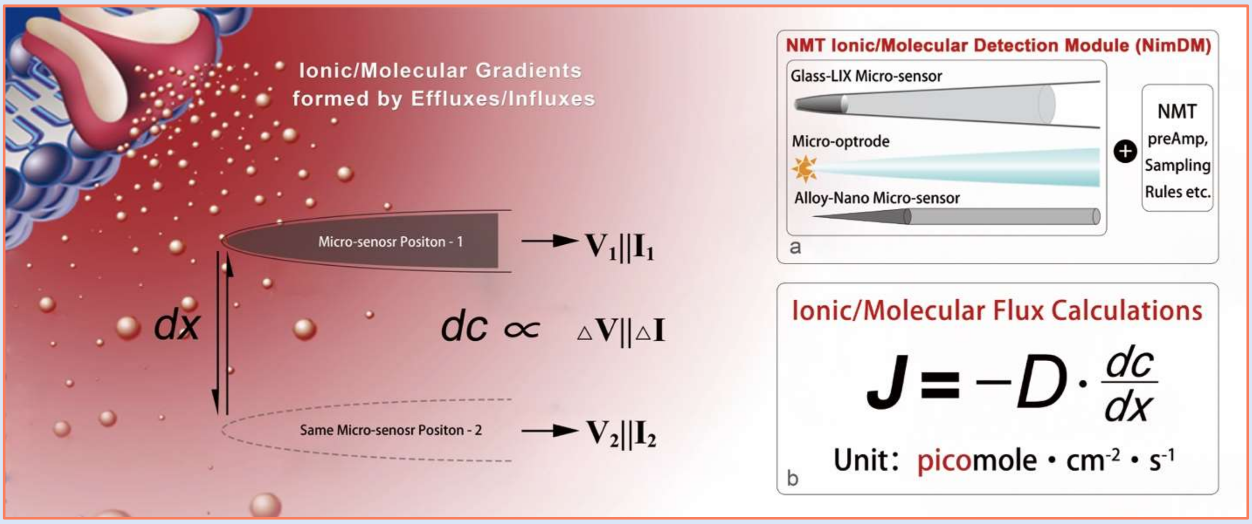


Figure 1. Working Principle of NMT. Diagram of NMT working theory. A NMT ionic/molecular micro-sensor undergoes two-point measurement at a known distance *dx* in the concentration gradient of the ions/molecules obtaining voltages $V_1||I_1$ and $V_2||I_2$ respectively. The concentration difference *dc* between the two points is proportional to $_{\triangle}V||_{\triangle}I$ derived from $V_1||I_1$ and $V_2||I_2$. **a**: fluorescent dyes/optical fibers, nano-carbon filaments, enzyme electrodes, fabricated metals/alloys, etc. can all be used to detect ions/molecules. **b:** Fick's first law of diffusion is adopted to calculate ionic/molecular fluxes, where *D* is the diffusion coefficient of ions/molecules (unit: cm^2 • s^{-1}).

2.1.2 Fluxes & voltage differences

The voltage differences stemming from the two-point measurement scheme of NMT is a key to understand how NMT works and why it can overcome the drifts of micro-sensor voltage readings **(Figure 2.)**. For instance, the voltage readings V_1 from 'Position-1' (solid line) will keep drifting even the measured ion's concentration is not changing at all. This drift makes it impossible to detect small concentration changes which forms the ionic and/or molecular concentration gradient resulted from their cross-membrane transportations. However, by using the same NMT micro-sensor with the same drifting pattern in 'Position-2', dashed lines in Figure Voltage Differences Cancel out NMT Micro-sensor Electrical Drifts, ΔV (V_1-V_2) will cancel out these drifts. Namely, if ΔV is not '0', then there is an ionic concentration gradient, and the magnitude of flux is directly proportional to the $|\Delta V|$. Moreover, the '+' and '-' signs of ΔV determines the motion directions, efflux or influx of the ions/molecules we are measuring respectively.

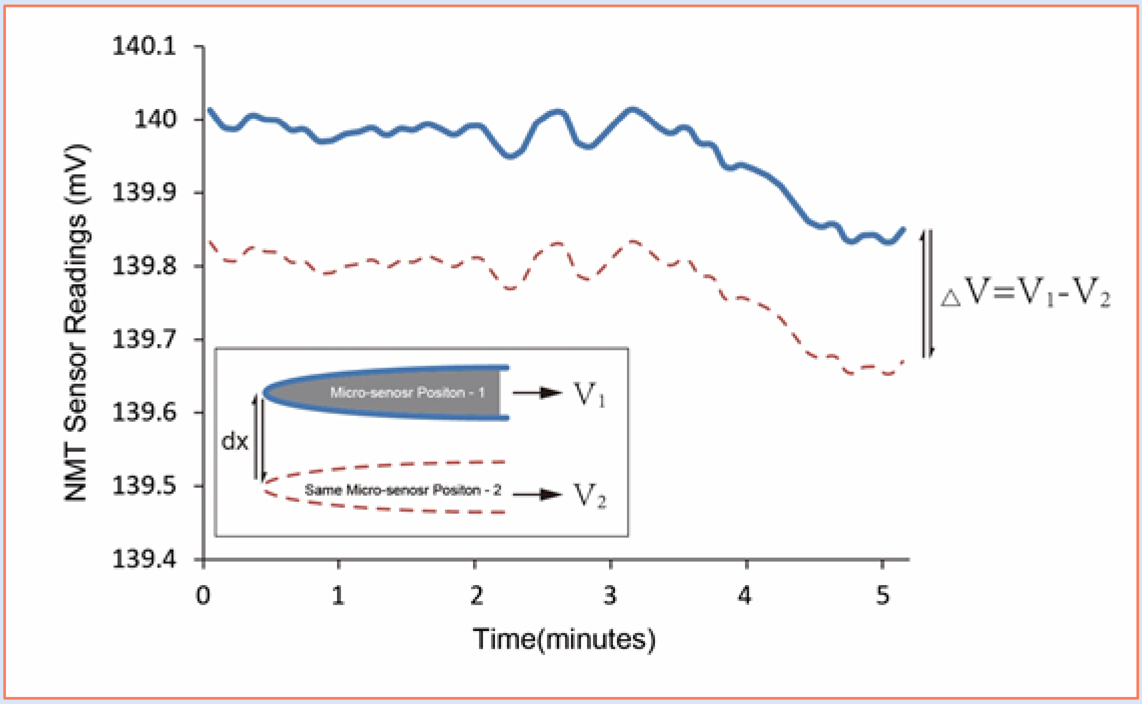


Figure 2. Voltage Differences Cancel out NMT Micro-sensor Electrical Drifts. Relationship between ΔV and drift readings of NMT sensors. A real time course plot of a NMT sensor millivolts reading from 'Position-1 (solid line)' to 'Position-2 (dashed line)'. V_1 from 'Position-1' and V_2 from 'Positon-2' will be read constantly in a 'back-forth' vibrating pattern of movement around 0.3Hz frequency. ΔV is calculated at the end of each cycle of sensor movement. **Insert:** shows the same sensor moving from Position-1 to Position-2 generating V_1 and V_2 respectively at distance dx.