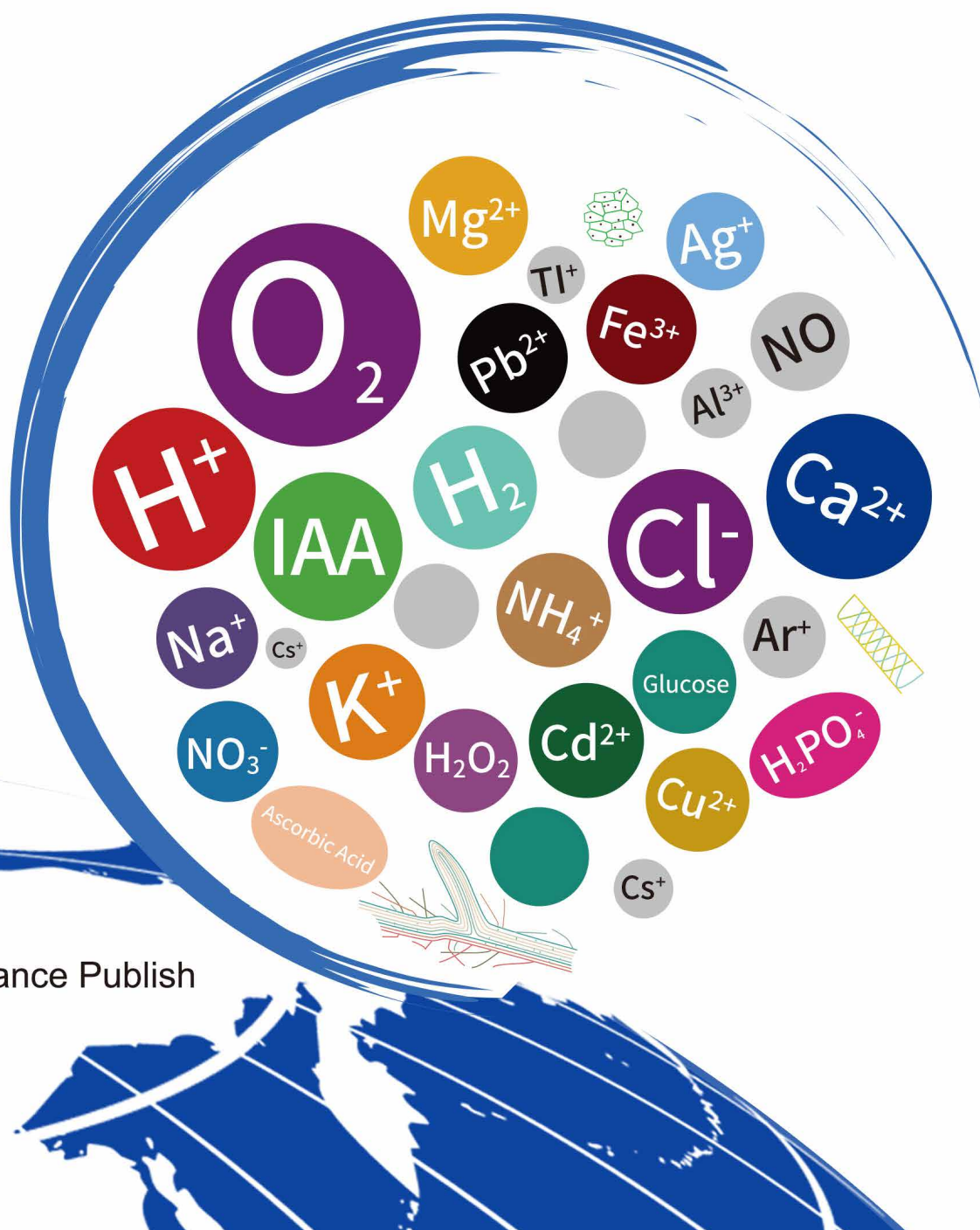


NMT

March 2023 Vol. 1 No.2

COMMUNICATIONS



NMT International Alliance Publish

ISSN 2834-5363

Dear editorial department of 《NMT Communication》 :

Thank you for telling me the good news of the founding of 《NMT Communication》 , I'm so delighted!



D. Marshall Porterfield PhD

Professor of Biological Engineering

Purdue University

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I would like to extend my warm congratulations on the founding of your journal! At the same time, I would like to extend my sincere greetings to all staff of the editorial department!

As an honorary editor of 《NMT Communication》 , I will assume the responsibility and mission of editing work, adhering to the actual needs of scientific researchers, and focusing closely on the content and application of Non-invasive Micro-test Technology (NMT) to provide a popular scientific platform of NMT knowledge and experience.

Please accept my best wishes for your future success, good fortune, and the attainment of new triumphs.

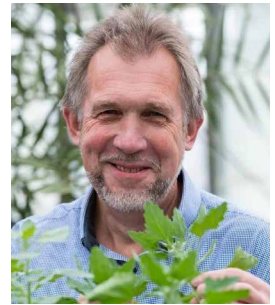
Dear editorial department of 《NMT Communication》 :

My seasonal greetings and sincere congratulations with the launch of this journal.

Over the last few decades non-invasive ion flux measurements (such as NMT technology) has transformed from being an occasional tool used by few selected researchers to a broadly accepted technological platform that allows to quantify activity of various membrane transporters in living organisms with very high spatial and temporal resolution.

The range of its application is extremely broad, from plant and animal physiology to environmental and food microbiology, marine biology, environmental science, and medicine. The technique is unique in terms that it can allow in situ quantification of electrogenically neutral transporters and is a nice complementation to (Nobel-prize winning) patch-clamp technology.

The launch of this journal will establish an efficient and timely platform for all NMT users that allow unrestricted exchange of ideas and foster fruitful collaborations between NMT users. I am looking forward to hear some exciting news about most recent developments in the field and would like to congratulate once again Jeff Xu and his team with achieving this milestone.



Professor Sergey Shabala, PhD

Professor in Plant Physiology

University of TASMANIA

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Cover Story

Editor's note:

The cover story of this issue of NMT Newsletter is from the article titled "Science and Technology" or "Technology"? Are we wrong. In this article, Professor Xu reflected on history, culture and education, and drew the conclusion that our Chinese people do not pay enough attention to 'technology' at present, and pointed out that although we are still weak in 'technology', every time we encounter a national crisis, it can stimulate our Chinese people's innovation ability and accelerate the improvement of our self-correction and self-innovation ability. I believe that in the near future, we Chinese people can not only achieve 'technological' innovation, but also step by step to the forefront of world scientific and technological innovation!

About the author:



Yue Xu , inventor of Non-invasive Micro-test Technology, founder of living functional genomics, practitioner of scientific and technological achievements transformation, advocate of international scientific cooperation, former senior researcher of NASA, president of Younger USA, LLC, chairman and CEO of Xuyue (Beijing) Technology Co., Ltd., chairman of Zhongguancun Xuyue Non-invasive Micro-test Technology Industry Alliance, and initiator and chairman of International NMT Alliance.

Younger USA, LLC was founded in 2001. In 2004, inspired by the older generation of scientists, such as the tutor of the Chinese Academy of Sciences, Kuang Tingyun, Academician Yang Fuyu and Professor Lin Kechun of Peking University, he resigned from the position of senior researcher of NASA in 2005 and returned to China to establish Xuyue (Beijing) Technology Co., Ltd., introducing the concept of oscillation electrode of American MBL scientist Lionel Jaffe into China, Under the guidance of the policy of "introduction, digestion, absorption and re-innovation" of the government's science and technology department and the support of the initial funds for entrepreneurship, led the Xuyue team, together with more than 2000 scientific researchers nationwide, through more than 20 years of unremitting efforts and concentrated research from 2001 to 2022, to forge the non-damage micro-measurement technology (NMT) and its series of application equipment with Chinese independent intellectual property rights.

In the process of completing the commercialization and industrialization of NMT in the field of scientific research,

(Editor in charge: Xuefei Li)

Date of receipt: February 26, 2022; Received on: February 28, 2022

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it helped domestic and foreign scholars to successfully apply NMT to 146 core articles in Chinese, 502 SCI articles, including 19 articles in top journals, such as CELL, NATURE, SCIENCE, etc., with a total impact factor of 2123. NMT scientific research equipment was exported to the University of Zurich in Switzerland, Europe, in 2020, completing the leap from technology to lead. In the past ten years, the transformation process of NMT's achievements in medical, health, environment, food, epidemic prevention, new materials, new energy, modern agriculture and other fields of people's livelihood has been gradually started, and gratifying progress has been made. Therefore, in June 2021, it passed the international leading level review of the institutions recognized by the Ministry of Science and Technology, and on this basis, it launched the establishment of the "International NMT Alliance" in 2022, Build the Chinese NMT team into a Non-invasive Micro-test Technology innovation force with international influence.

Graduation experience

Summary of learning experience and personal experience of non-injury advanced seminar

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Abstract: Non-invasive micro-measurement is an important technology in electrophysiological research. It can obtain real-time physiological and ecological information of samples in a nondestructive manner, and has important biological significance for studying the laws of life activities and environmental impact. After learning the non-invasive technology, I carried out many related experiments, tested a variety of samples, and had some own experience and understanding. It happened that Xuyue Company held an advanced seminar on the application of Non-invasive Micro-test Technology, and invited many famous teachers to explain the application of non-invasive technology. Therefore, this article briefly introduces the Non-invasive Micro-test Technology and its development in China, the learning experience in the seminar and my personal experience in the non-invasive experiment.

Key words: non-injury, electrophysiology, plant adversity

I. Introduction to Non-damage Micro-measurement Technology and Its Development in China

Non-invasive micro-test technology (NMT) was proposed in 1974 by Lionel F. Jaffe, a neuroscientist at the Woods Hole Marine Biology Laboratory in the United States. Non-invasive micro-test technology is composed of scanning ion selective sensor technology (SIET), micro-sensor ionization current technology (MIFE), self-reference polarographic sensor technology (SERP), scanning polarographic sensor technology (SPET), scanning vibration sensor

technology (SVET) Self-reference ion selective sensor technology (SERIS) and other technologies. It is a technology to study the physiological function of living materials. It can detect the flow rate (flow rate and direction) of molecules/ions entering and leaving the living body without damaging the sample. The unit is $\text{pmol} \cdot \text{cm}^{-2} \cdot \text{s}^{-1}$, which is called flow rate (Flux). It is a non-contact method with ultra-high sensitivity. Because of its characteristics of in vivo detection, non-invasive, high resolution and dynamic real-time,

Date of receipt: December 4, 2022; Received on: December 4, 2022

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it is often used to explore physiological characteristics that are difficult to measure by other technologies. It is one of the best tools for physiological function research at present, and has been widely used in medicine, botany, zoology, microbiology, agricultural science, pharmacy, environmental science and other fields. At the same time, NMT is also the technical basis of dynamic ionics. After years of development, it has become a mature experimental system. He has published a large number of relevant scientific research achievements in the top international magazines Science, Nature, PNAS, Plant Cell, Environmental Science&Technology, etc. The in vivo, dynamic and real-time measurement methods of Non-invasive Micro-test Technology, as well as high resolution and high sensitivity, help human to carry out research in the field of physiological function, and have been applied in many disciplines.

NMT technology has been developed in China for many years. Since Mr. Xu Yue founded the American Younger Company in Massachusetts in 2001, NMT has entered the stage of commercialization. Xuyue (Beijing) Science and Technology Co., Ltd. was established in Zhongguancun, Beijing, in 2005. The academic expert advisory group was composed of Kuang Tingyun, academician of the Institute of Botany of the Chinese Academy of Sciences, Yang Fuyu, academician of the Institute of Biophysics of the Chinese Academy of Sciences, and Lin Kechun, professor of Peking University, and became a strategic partner with the United States Younger Corporation. NMT officially entered China. In 2014,

the "Second Non-damage Micro-measurement Technology Seminar" was held in Beijing, and the "NMT International Standardization Committee" was established. In 2015, Xuyue (Beijing) Biological Function Research Institute was established in Beijing. In 2015, the "Zhongguancun Non-injury Micro-measurement Technology Industry Alliance" was established by Xuyue (Beijing) Biological Function Research Institute. The alliance is committed to the industrialization development of NMT. At present, more than 20 scientific research institutions and enterprises have joined the alliance.

II. Learning experience of the seminar

Many teachers in the seminar shared their NMT-related experimental research, which benefited me a lot. The following is my own learning experience:

Professor Zhuo Renying mainly taught the screening and functional analysis of the key genes for cadmium enrichment in *Sedum paragonum*. 48 genes related to cadmium tolerance were screened by yeast transformation system. It is predicted that these genes may participate in MAPKs-mediated signal transduction, calcium ion signal, reactive oxygen species signal and hormone signal. The SaHsfA4ch gene can improve the cadmium ion tolerance of yeast strain Δ ycf1. Compared with the control, the overexpression of SaHsfA4ch was detected by non-invasive micro-measurement. The cadmium ion influx increased, which promoted the uptake of cadmium ion by plants, enhanced the tolerance of Arabidopsis

to cadmium ion, and increased the activity of active oxygen scavenging enzymes. SaHsfA4ch gene was mainly expressed in vascular tissue, flowers and stems, and was significantly induced under cadmium stress. Then we found three downstream genes regulated by SaHsfA4ch gene: SpPDR12, SpPCS2 and SpHsfA4c. Through EMSA and LUC activity detection, it was found that SaHsfA4ch could bind to the HSE element of the SpAPX2 gene promoter. SaHsfA4ch gene significantly improved the cadmium tolerance of transgenic *Sedum alfredii*. Professor Zhuo's research found the cadmium ion tolerance gene in *Sedum*, studied its main function, and applied it to the control of heavy metals in combination with practice, which is a very meaningful work.

Professor Sun Jian mainly explained the molecular mechanism of carbon point regulating the stress resistance of sweet potato. Carbon dots have the ability to promote electron transfer, photosynthesis and ROS removal. The carbon dots synthesized by *salvia miltiorrhiza* powder can induce the hyperpolarization of the root cell membrane of sweet potato and the calcium ion influx independent of ROS, and have the same effect in *Arabidopsis*, tobacco, rice and other crops. It is found that the calcium ion influx induced by carbon dots is through the hydroxyl and carboxyl functional groups on the surface. The increase of potassium ion and ammonium ion influx in root cells caused by carbon dots proves that carbon dots activate the hyperpolarized non-selective cation channel, which is presumed to be a cyclic nucleotide channel. Then it was detected that carbon point could increase the content of cAMP and cGMP in root

cells of sweet potato. After that, it was found that CNGC2/4/19/20 mediated the increase of calcium ion influx and cytoplasmic calcium ion in *Arabidopsis* root cells induced by carbon dots. The calcium ion influx induced by carbon dots in *Arabidopsis* depends on the lectin receptor kinase LORE and LERCK-1.8. Applying carbon dots to hydroponic seedlings could enhance the outflow of calcium and sodium ions from root cells of sweet potato under salt stress. The carbon point increases the sodium ion outflow from *Arabidopsis* roots under salt stress, which depends on CNGC2. Carbon spots can inhibit the absorption of sodium ions and reduce the accumulation of sodium ions at the cell and whole plant level in the root mature zone. The carbon spot enhanced the expression of root calcium signal related genes and the tolerance of sweet potato to nutritional stress. Mr. Sun studied the role of nanomaterials in the resistance of sweet potato to adversity. Nanotechnology, as an emerging technology, has good prospects for development. In the study, NMT was used to detect the ion flow rate of sweet potato after the application of carbon point, and intuitive and clear experimental data were obtained, which can well reflect the treatment effect. The research content is relatively in-depth, and the functional group and action channel of carbon point have been found, which plays an important role in the research of sweet potato stress resistance.

Professor Zhang Jinlin mainly studies the application of NMT in the study of salt tolerance mechanism and salt tolerance germplasm screening of gramineous forage. It was found that the salt vesicles on the stem and leaf surface of *Atriplex tetraptera* played an

important role in salt resistance and drought resistance. A variety of plants were selected to detect the ratio of sodium and potassium ions in their soil, and it was found that the ratio of sodium and potassium ions in large areas was the highest. By comparing the content of sodium ion in wheat with that in wheat, it was found that the content of potassium ion was significantly lower than that in wheat. This shows that the alkali grass absorbs a lot of potassium ions, but reduces the absorption of sodium ions. The amount of salt secreted on the surface of leaves was measured by washing liquid experiment, and it was found that salt secretion was not the main reason for the salt tolerance of *Puccinia arundinacea*. After that, using isotope labeling method, it was found that the sodium ion influx rate of the root system of *Puccinia arundinacea* was lower than that of wheat. In the case of potassium ion starvation and high concentration of sodium ion, sodium ion efflux and hydrogen ion efflux decrease. This indicates that the sodium hydrogen reverse transporter consumes more hydrogen. Then we studied the genes related to sodium ion transport, and found that under potassium starvation, the expression of AKT1 gene was higher, which participated in the absorption of high-affinity potassium ions; Under the condition of high salt and potassium starvation, HKT1:4 and HKT1:5 were expressed in xylem parenchyma cells, and sodium ions were excreted through the plastid pathway. Mr. Zhang studied the salt-tolerance mechanism of saline-tolerant plant and found the related genes of sodium ion transport, and studied their functions in sodium ion transport. As an important function of plant survival under adverse

conditions, salt tolerance of plants has important research significance, which can effectively improve the situation of plants that are difficult to survive in high saline-alkali soil.

III. Non-injury personal experience and experimental scheme setting

I gradually learned the convenience of this technology in the process of using the non-invasive system to test the ion flow rate of the sample. At the same time, I also have some personal experience as follows:

1. Solution configuration needs to be accurate. The test solution, calibration solution and filling solution all play a very important role in the experiment process, and the ion concentration of the test solution and calibration solution is very low. Therefore, it is necessary to prepare the mother solution first. The mother solution can be configured with 50 ml, and the weighing powder is greater than 0.1 g, which will be more accurate. During dilution, it can be diluted 10 times or 100 times at a time. If the dilution ratio is too high at a time, the ion concentration will also be inaccurate. Refer to the NISC test solution standard for the configuration of the test solution. The ion concentration can be adjusted appropriately, but some necessary conditions should be met: the test solution should contain the ions to be tested; The ion concentration of the test solution should not be too high or too low, otherwise the detection sensitivity will be affected; The test solution needs to maintain a certain PH value to maintain the stability of the

sample (the cell sample also needs to adjust the osmotic pressure); The high concentration calibration solution and the low concentration calibration solution are only different from the concentration of the ions to be measured, and the rest are consistent with the test solution. The concentration difference of the ions to be measured should be 10 times; When adjusting the PH value, the solution containing the ions to be measured cannot be used, but Tris and MES can be used.

2. There are many kinds of samples that can be tested without damage. The plant samples can be tested by using water culture, soil culture and medium culture, but the most suitable one is water culture, so that the surface is clean. When using soil culture and medium culture, it is better to brush the test part gently with a fine brush to remove the impurities on the surface, and then soak in the balance solution before testing.

3. Production of flow sensor. First, take a sensor and inject 1 cm of filling liquid. The position should not be too far from the tip, otherwise it is not easy to squeeze the filling liquid to the opening. Then connect the sensor to the three-way valve, push the syringe to make the filling liquid rise to the tip and flow out 1 drop, and gently blow off the flow out of the liquid drop with your mouth against the sensor. Dip the LIX holder in the LIX and fix it on the micromanipulation table. Slowly push the pressure adjusting device to make the LIX holder form a convex surface and adjust the focal length to make the picture clear. Then slowly adjust the position of the sensor so that the sensor tip is inserted into the LIX holder convex liquid level to

make the LIX flow into the sensor. Gently push the three-way valve back and forth several times, and then adjust the LIX liquid column to the appropriate length to separate the flow sensor from the LIX liquid level.

4. The flow sensor shall be adjusted manually and automatically. During the test, the tip of the velocity sensor should be immersed in the liquid level, not only a little, which will affect the detection effect, but also not too deep, which may touch the bottom of the petri dish and cause the tip to break. Therefore, when adjusting the position of the sensor, it is better to put the sensor down to the liquid level or close to the liquid level, and then rotate the Z-axis knob to make the sensor slowly lower. After the tip is immersed in the liquid level, properly adjust the X-axis and Y-axis to make the sensor tip in the middle of the picture. Finally, turn on the automatic switch, use the automatic adjustment to make the tip clear, and observe whether the tip is complete and not broken. The calibration can be carried out according to the sequence of high concentration calibration solution, low concentration calibration solution and test solution during calibration. The next calibration solution can be replaced after each calibration is stable.

5. Some reasons for the calibration error of the calibration solution: (1) The detection values of the three solutions are very low and there is no difference. This may be because the machine is not turned on. Check whether the switch button is turned on; (2) The detection values of the three solutions are very high, which is much higher than the normal value.

This may be because the circuit is not closed, and whether the reference electrode is put into the liquid level is detected; (3) The measured values of the three solutions are within a reasonable range and the three concentrations have gradient differences, but the slope is incorrect. This may be due to the problem of the calibration solution configuration. You can dilute the high concentration calibration solution by 10 times for testing. If it can be calibrated, it means that the dilution with the mother solution is not accurate. If it can not be calibrated, it means that there are other problems, such as adding too much potassium ion to the detection of sodium ion, which affects the detection.

The design of experimental scheme is an important part of NMT test. The following are some experimental schemes I have done myself:

1. Potassium ion flow in plants under salt stress.

Potassium ion is a major element necessary for plant growth, and plays an important role in plant nutrition, turgor regulation and osmotic balance. High salinity and alkalinity will form high osmotic pressure, which will make plants lose water and affect the normal growth of plants. *Arabidopsis thaliana* is used as the test sample. Take plants with similar growth for 7 days and put them into 100 mM NaCl solution for treatment for 24 hours, and use the solution without NaCl as the control. Soak the treated material in the test solution for 30 min, then replace it with a new test solution, and fix the sample with filter paper strips and resin blocks. The velocity sensor detects the velocity of potassium ion at the root tip.

2. Changes of calcium ion flow rate in plant roots under low temperature treatment. Calcium is the second messenger of plant signal transduction and plays an important role in signal transduction and regulation of plant growth and development. The normal development of rice will be affected by low temperature chilling injury, which will lead to yield reduction. Therefore, it is of great significance to study the changes of calcium ion transport in rice under low temperature for the study of low temperature stimulation. In the experiment, put the rice sample into the test solution for balance for 30 minutes, and then replace it with a new test solution for test for 5 minutes. After the data is stable, add crushed ice into the outer culture dish to create low temperature conditions, and detect the flow of calcium ions during low temperature treatment.

3. Absorption of plant roots after heavy metal treatment. Cadmium is a heavy metal element with strong toxicity and belongs to category 1 carcinogen. If a certain concentration of cadmium is accumulated in plants, it will affect the growth and development of plants. Cadmium stress usually stimulates the growth of plant seedlings at low concentrations and inhibits the growth of seedlings at high concentrations. *Arabidopsis thaliana* was used as the research material and put into the test solution containing 0.05 mM cadmium to balance for 30 minutes. Replace the test solution with a new one, fix the sample, and measure the cadmium ion flow rate at the root.

After years of development, Non-invasive Micro-test Technology has become an important method in electrophysiological experiments and has irreplaceable advantages. The application of Non-invasive Micro-test Technology has greatly promoted the research progress of ionic molecular flow rate signals. I am deeply impressed by the sense of social responsibility of Xuyue Company in organizing the seminar to promote the Non-invasive Micro-test Technology, and I am also honored to participate in the study of this seminar, which has taught me a lot about NMT. It is hoped that the non-injury technology can be extended to more scientific research institutions in the future to serve the scientific research work.

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(Editor in charge: Xuefei Li)

Research on the mechanism of plant stress resistance assisted by Non-invasive Micro-test Technology

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Abstract: A gene can change the fate of a country, a seed can change the future of a nation, and a new technology will certainly promote the leapfrog development of related disciplines. We are in a good era of doing scientific research. In this era, new disciplines and new technological methods emerge in an endless stream, which will escort us to carry out scientific research. Non-invasive micro-test technology (NMT) can detect the real-time dynamic changes of various ions or molecules in living plants. At present, this technology has reached the international leading level. It has shown great application potential in the fields of plant nutrition, saline-alkali stress and heavy metal stress response, providing an ideal solution for the study of plant response to environmental dynamics. This paper briefly reviewed the development of NMT, and the main teaching contents of relevant experts in the field of plants under the 2022 Advanced Seminar on the Application of Non-injury Micro-measurement Technology. At the same time, combined with my research work, I designed relevant topics, and put forward suggestions for the development of NMT.

Key words: Non-invasive Micro-test Technology, plant stress, salt stress, heavy metal, physiological and biochemical

I. Introduction to Non-damage Micro-measurement Technology and Its Development in China

Non-invasive micro-test technology (NMT) was first proposed by American scientist Lionel F. Jaffe, and then gradually applied to solve many key scientific problems through continuous exploration and development (Liu et al., 2022). Professor Xu Yue is a pioneer and leader in the localization of NMT in China. He has guided the development of NMT in China for nearly 20 years, and promoted the

application of a vibrant and vigorous technology. At present, NMT technology has reached the international leading level, has obtained a number of patent technologies, and has been widely used in life science, material science, earth science, national economy and people's livelihood and other fields.

Date of receipt: December 4, 2022; Received on: December 4, 2022

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At present, NMT can be used to measure the real-time changes of sodium (Na^+), potassium (K^+), calcium (Ca^{2+}), magnesium (Mg^{2+}), ammonium (NH_4^+), cadmium (Cd^{2+}), lead (Pb^{2+}), copper (Cu^{2+}) and other heavy metal ions between living tissues, cells and the internal/external environment, as well as multiple ion or molecular indicators such as NO_3^- , Cl^- , O_2 and IAA (Liu et al., 2022). These indicators can reflect the dynamic response of living organisms to the external environment, providing strong physiological evidence for revealing the physiological mechanism.

Food security and environmental pollution prevention and control issues related to the national economy and the people's livelihood have aroused widespread public concern. The Zhongguancun NMT Industrial Alliance pays attention to the application and promotion of theory and technology, and actively serves the majority of scientific researchers. In October 2022, the Alliance organized the 2022 Advanced Seminar on the Application of Non-injury Micro-measurement Technology • Plant Field, aiming to cultivate a group of scientific researchers who are familiar with and use NMT, which will certainly set off a research upsurge of NMT technology in the three "pillars" of plants (salt and alkali stress, heavy metals and plant nutrition). This seminar invited 12 experts, scholars and application engineers from front-line scientific research to give lectures to the students. The detailed scientific research data support a rich scientific story. We are deeply impressed by the rigor and difficulty of scientists. Every article we read should be respected

and feared. By participating in this seminar, we can have a deeper understanding of the application of NMT in plant nutrition, plant nanobiology, salt stress and heavy metal stress, education and teaching, and seed industry.

II. Application of Non-invasive Micro-test Technology in plant nutrition, nanobiology and stress response research

Root is an important organ for plants to absorb nutrients and water directly. Root configuration, including root length, number of lateral roots, number of root hairs, root orientation or water orientation, is the key factor determining nutrient and water absorption. In the context of climate change, the global arid and semi-arid regions will be further expanded (Huang et al., 2016). Therefore, analyzing the regulation mechanism of plant response to water will help improve and improve the utilization efficiency of water resources and nutrients. Professor Xu Weifeng's team has established a system for studying the hydrophilicity of roots. The research found that the hydrophilicity of plants is characterized by the ability of roots to anchor water and grow towards the soil with water under the stress of drought and water shortage. It also found that brassinolide BR can promote the hydrophilicity of roots by regulating the root proton flow signal (Miao et al., 2018). In this study, the dynamic changes of H^+ , Ca^{2+} and H_2O_2 were detected by NMT. In addition, the hydrophilicity of root is also closely related to the formation of root

sheath. The root sheath is an adaptive structure formed by the mutual cementation and twining of the mucus secreted by the rhizosphere soil particles and roots, the mucus secreted by the rhizosphere microorganisms and the root hairs, which is particularly important for plant drought resistance. The research found that the root sheath is the "highway" for the nutrient and water absorption of crops. The endophytic fungus *Pyrosporium indicus* under moderate irrigation can promote the construction of rice root sheath, providing important reference value for the efficient use of water by crops (Zhang et al., 2020; Xu et al., 2022).

Nanotechnology has been recognized by the European Commission as one of the six most promising key technologies in the 21st century, and has been widely used in biomedicine, environment, food and agriculture and other fields (Salata, 2004). The negative or positive effects of engineered nanomaterials (EMs) on plant growth and development are attributed to their physical and chemical properties, application concentration, etc. Engineering nanomaterials have great potential in improving plant saline-alkali stress and heavy metal tolerance (Wan et al., 2020; Zhao et al., 2020). Carbon-based nanomaterials, including carbon dots (CDs), single-walled carbon nanotube angles (SWCNHs), single-walled carbon nanotubes (SWCNTs), multi-walled carbon nanotubes (MWCNTs), graphene oxide and fullerene (C₆₀), are new engineering nanomaterials with broad application prospects (Lahiani et al., 2015). Biomass derived carbon dots are typical zero-dimensional carbon-based nanomaterials with good water solubility,

biocompatibility and low toxicity. Sun Jian's team found that lectin receptor kinase LecRKs can sense the -OH and COOH functional groups on the surface of the carbon point of *Salvia miltiorrhiza*, thus promoting the accumulation of cAMP/cGMP and activating CNGCs. In this case, cytoplasmic Ca²⁺ mobilization does not depend on ROS signal. The study also found that under the stress of NaCl, low K⁺ and low Fe²⁺, CDs promote the concentration of Ca²⁺ in the root cytoplasm to increase, and then affect the absorption of K⁺, Na⁺ and Fe²⁺, thus improving the adaptability of plants to these stresses (Li et al., 2022). The above research shows that engineering carbon nanomaterials such as SWCNHs and CDs have important application prospects in the fields of sustainable agricultural development and resources and environment.

Saline-alkali stress is one of the main abiotic stresses that endanger plant growth. China's saline-alkali land is more than 500 million mu, and it is estimated that more than 50% of the cultivated land will be saline-alkali by 2050. The red line of 1.8 billion mu of arable land should be maintained, and at the same time, 500 million mu of saline-alkali land should be fully developed and utilized. Therefore, it is necessary to change the traditional conventional crop breeding method to carry out the research on the saline-alkali tolerance mechanism of plants, especially crops. Cultivating saline-alkali tolerant crops will provide important support for China's granary and rice bowl. Saline-alkali tolerant plants adopt different strategies such as salt rejection and salt secretion to reduce salt absorption and thus reduce toxicity. The dynamic

detection of Na^+/K^+ ion flow by NMT provides strong support for plants to respond to saline-alkali stress. Professor Zhang Jinlin's team found that the toxicity of Na^+ was reduced by reducing the inflow of Na^+ in the root system; Further research found that the salt tolerance of *Puccinellia parviflora* was affected by *PtSOS1* and *PtHKT5; 1* and other ion transporters/channels *PutHKT1; 4*, *PutHKT2; 1* and *PutAKT1* (Han et al., 2022). The team also used NMT to screen the salt tolerance of oat varieties, revealing the differential mechanism of *AsSOS1* regulating their response to salt stress. Salt tolerant varieties have strong root Na^+ absorption control ability, root Na^+ elimination ability and K^+ retention ability (Zhang et al., 2022).

Heavy metal stress seriously affects the growth and development of plants, especially the crop yield and products, and endangers food security and human health (Zhao et al., 2021). Cd is the primary pollutant of soil pollution in China, and the excess rate of Cd in soil is as high as 7%. The prevention and control of heavy metal pollution is a major strategic demand of China. Physical remediation, chemical remediation, phytoremediation and other technologies have been widely used in the remediation of soil contaminated by heavy metals. Poplar is a fast-growing tree species with developed roots, large aboveground biomass, strong adaptability and wide distribution, and high total Cd accumulation. It has great potential in the application of heavy metal Cd remediation. Professor Luo Zhibin's team has long been committed to the research on the regulation mechanism of poplar Cd enrichment. The hybrid poplar (*Populus tremula* x *P. alba*) is a poplar variety with strong Cd accumulation

ability selected by his team (He et al., 2013). The research found that leaves and bark are the main parts of poplar Cd accumulation. In addition, the top of root tip (0-900 μm) is the main segment of Cd absorption, which may be related to the development degree of root tip tissue Kjeldahl zone. The length of this region is shorter than that of the root elongation region and the mature region. Even though the Cd absorption velocity is fast, the accumulation of Cd in other regions may also play an important role in the total Cd accumulation. Therefore, it is necessary to detect the dynamic changes of Cd uptake in poplar from time and space scales, and explore the different mechanisms of Cd uptake in different regions of poplar roots. They further found that glutathione and ectomycorrhizal fungi were involved in regulating the tolerance of poplar to Cd stress (Ma et al., 2014; Shi et al., 2019). The content of Cd in poplar litter is significantly lower than that in fresh leaves, which suggests that Cd is activated and transferred in poplar leaves, but the specific mechanism remains to be further revealed.

Obviously, the wide recognition and application of NMT cannot be separated from the dutiful researchers and engineers of the Zhongguancun NMT Industry Alliance. Write NMT into the experimental guide of textbooks, let more scientific research institutes and universities understand and use NMT, which plays an important role in promoting NMT, and is also an important measure to implement the national strategy of rejuvenating the country through science and technology. In addition, NMT has also developed

a user-friendly guide for visual operation, which provides a convenient channel for new users to learn about NMT. I believe that in the near future, NMT will shine brilliantly in education, scientific research and other fields.

III. Take advantage of NMT to inject new impetus into the research

My research direction is crop nutrition and environmental adaptation, mainly focusing on the physiological and molecular mechanisms of crop response to nutrition and environmental changes, especially the mechanism analysis of upland rice adaptation to terrestrial environment, including low water germination of seeds, emergence of seedlings after germination, response to high or low temperature during growth period, grain filling and endosperm filling, and rice quality. Upland rice, also known as upland rice, is the xerophytic ecological type of Asian cultivated rice. It is planted in an aerated and aerobic dry land environment. The dry direct seeding cultivation of upland rice is faced with complex dry land environment. In production practice, problems such as difficult seedling emergence, uneven seedling emergence, and late lodging are often encountered. Cultivating drought-resistant and water-saving rice varieties and developing upland rice are important development directions for rice production under the situation of water shortage, labor shortage and seasonal drought. Systematic screening and evaluation of upland rice germplasm resources adapted to the terrestrial environment is the key to the development

of upland rice.

We will further enrich the content of the research project and improve the quality of the published scientific research results by applying the nondestructive micro-measurement technology. With the help of NMT (SVS-100 system), the vitality of different upland rice germplasm under drought stress was studied using more than 2500 upland rice germplasm resources collected by our research team in the early stage. The main contents include the following aspects: (1) The oxygen consumption during seed germination was detected by simulating different concentration gradients of drought stress, such as 10%, 15%, 20% PEG6000 treatment; (2) Storage of seeds with different responses, including soluble sugar, total protein, etc., and α Amylase and β The physiological and biochemical indexes such as amylase activity were measured to compare the physiological differences of different upland rice germplasm in response to drought stress; (3) Further carry out systematic evaluation of seedling growth indicators to evaluate the reliability and accuracy of NMT early rapid response identification results; (4) Through the construction of genetic population materials, the important candidate genes are precisely located and their regulatory mechanisms are analyzed. Through the above research, we hope to screen excellent upland rice germplasm resources with strong drought-tolerance ability, which can be used to guide subsequent pot or field drought-tolerance identification, analyze the physiological and molecular basis of upland rice to adapt to drought

stress, and provide important genetic materials and genetic resources for upland rice molecular design and breeding.

IV. Thinking and Prospect of Non-damage Micro-measurement Technology

After nearly 20 years of development and improvement, the nondestructive micro-measurement technology has become increasingly mature. The craftsman spirit of dedication, excellence, meticulousness and pursuit of excellence can constantly push our work to new heights. A new starting point and a new height. At present, the nondestructive micro-measurement technology has been recognized by the industry and scientific researchers, showing its unique charm and application prospects. Standing at the intersection of historical development, opportunities and challenges coexist. NMT can make more outstanding contributions to the progress of science and technology only by constantly seeking progress and excellence. In view of this, the following thoughts and suggestions are put forward, hoping to provide some reference for the future development of NMT.

First, work hard on the sensitivity of the detector. This requires optimizing and improving the threshold that each detector can detect, which will help reveal the precise mechanism of plant response to changes. As we all know, reactive oxygen species (ROS) is like a "double-edged sword" for plants. ROS with

appropriate concentration can be used as signal molecules to activate relevant signal pathways to enhance plant resistance; The excessive accumulation of ROS will cause damage to protein, nucleic acid and other substances and affect the normal growth and development of plants (Li and Kim, 2022; Zhao et al., 2022). The production and elimination of ROS in plants are in a dynamic balance, but due to the specificity of plants, the ability of each plant to maintain ROS balance is different. For specific plants, the threshold of role transformation of ROS as a signal molecule or stress signal has not been clearly revealed. Improving the sensitivity of NMT to detect O_2 will provide an answer to this question by detecting its dynamic changes in space.

The second is to expand the scope of detection of ions or molecules. There is still a lot of room for detecting ions or molecules, especially some multivalent elements and special functional organic small molecule compounds, such as metal elements iron (Liang, 2022), manganese (Zhang et al., 2021), plant hormones ethylene, abscisic acid (Yoshida et al., 2019), etc. These substances are essential for plant growth and development. At present, the indicators that can be detected are far lower than the number of important functional substances that plants respond to environmental changes. If NMT can detect different valence states of the same element, it will provide evidence for revealing the dynamics of the transformation of related elements in plants. At the same time, the interaction between plant hormones plays an important role in the regulation of

plant environmental adaptability. It is urgent to carry out the detection of relevant hormones, especially gas hormones, which will help clarify the function of hormone dynamic changes in plant response to environmental changes, and provide a basis for further development of genetic and molecular mechanism analysis.

The third is to develop new compact composite detectors. This kind of detector can be used to detect the dynamic changes of different target ions or molecules simultaneously and multi-dimensionally. Plant system is a complex organism, and its response to environmental change is a systemic and global response. If several ions or molecules of concern can be detected synchronously, the mechanism of its response will be better explained.

Fourth, improve the NMT sharing open platform and provide affordable testing services. Although there are more than 500 laboratories of more than 100 units in China with NMT equipment, the resources are relatively concentrated in developed regions, while the layout of northwest and southwest regions is relatively small. These areas are faced with more serious environmental problems such as saline-alkali stress, drought, heavy metal stress, etc. It is necessary to vigorously carry out research on the mechanism of the response of probiotic species to these stresses with the help of NMT. In addition, the cost of the current test is relatively high, which limits the space for researchers to use the NMT system to a certain extent. It is hoped that the Zhongguancun NMT

Industrial Alliance will be based on the long term and allow this technology to benefit more researchers in scientific research institutes and universities.

Epilogue

The Non-invasive Micro-test Technology provides us with a window to dynamically detect the response of plants to the dynamic changes of internal and external environment, and can accurately grasp the physiological response of plants in real time. It also injects new vitality into the screening and identification of excellent germplasm resources and functional gene analysis. It is hoped that NMT people and NMT colleagues will continue to innovate, pursue excellence, maintain the international leading sustainable power, and let NMT, the key core technology, play an increasingly important role in China's scientific and technological self-reliance.

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(Editor in charge: Xuefei Li)

Kehai Imagination

'Science & Technology' or 'Technology & science'? Are we wrong?!

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A Chinese NMT person said well: "Science and technology are the relationship between fish and water!"

Just as the NMT technology has promoted the progress of many disciplines in science, the development of these disciplines and theories in turn has put forward many innovative requirements for NMT technology.

However, from the Sino-Japanese War of 1894 and the subsequent Treaty of Shimonoseki to the Sino-US trade war, science and technology war, financial war and the American Chip Act against China this year since 2018, the Chinese nation has suffered from technological backwardness for more than 100 years, until today! This makes us NMT technicians feel deeply responsible and obligated. Think more about why our technology is backward? What did we do wrong?

Details:

"The devil is always hidden in the details!" is a phrase often spoken by technical people.

Of course, sometimes people who engage in technology will also go crazy. The following details

of life and their associations are somewhat far-fetched and even childish. But for the sake of the country's scientific and technological progress, what's the harm of being generous?!

There is a detail in Chinese and English translation. I wonder if you have noticed it.

How to translate the sentence "My name is Yue Xu."?

The accepted answer seems to be: "My name is Xu Yue." We will naturally reverse the order of surname and first name.

So, how to translate "Science and technology need innovation."? "Science and technology need innovation." seems to be the prevailing answer.

However, we know that in English world culture, surname is more important than first name. That is to say, in culture, subconsciously, we may not understand that Westerners think technology should be more valued than science! Is this a "devil's detail" that we Chinese people ignore when introducing advanced

Date of receipt: 2022-12-29; Received on: February 29, 2022

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western technology?

Is this a possible reason why we have lagged behind western science and technology for a long time? We got the order wrong?!

History:

Since humans have mastered the technology of "drilling wood to make fire", they have enough energy to supply the human brain with "scientific" thinking.

The Chinese people have four major 'technology' inventions, and only then have the Chinese people become the thousand-year continuous civilization that is proud of the world.

Qin's military technology, such as 'Qin sword' and 'strong crossbow', helped it unify China.

From the Sino-Japanese Sino-Japanese War to the Sino-US War of Science and Technology.

From the 1898 Reform Movement to the May 4th Movement in 1919, many Chinese people and people with lofty ideals have been looking for the way to a powerful country. The May 4th spirit of "patriotism, progress, democracy and science" has inspired the national rejuvenation enthusiasm of generations of young people. To be sure, the slogan "science" put forward that year must contain the meaning of "technology", but does it also reveal that we do not pay enough attention to "technology"?

Is it that we have mistaken the priority of "science and technology"?!

Culture:

The Chinese culture is extensive and profound. The philosophical thoughts of Confucius, Laozi, Mencius, Mozi, Han Feizi and other schools of thought, as well as the military theory of Sun Tzu's Art of War, have had a significant impact on human civilization and development. However, is it just like Lao Tzu said, "misfortune depends on fortune, and fortune depends on misfortune. After seeing through the way of everything, the Chinese people, compared with other national cultures, are relatively lack of the desire to further understand nature, and thus lose the motivation of 'technology' innovation that can use or even change nature?

For example, the national treasure of traditional Chinese medicine, in addition to a few clear mechanisms of action like artemisinin, from the perspective of modern life science, we do not know why traditional Chinese medicine can cure diseases. Moreover, our culture always seems to say to us in the ear: "Thousands of years have proved that it can cure diseases, why should we have to figure out why?!"

We believe that traditional Chinese medicine is a science, but its development is relatively stagnant, and today China's development is still blocked by others' 'technology'!

Isn't it because we don't pay enough attention to 'technology' in our culture?!

Education:

Criticized for many years, but our children 'read dead books, read dead books!' It is still a common phenomenon.

In addition to memorizing books and some scientific concepts and theories, our students usually have poor practical skills. The practical ability is closely related to the 'technological' innovation ability.

A foreign professor's evaluation of Chinese overseas students is right: "The basic knowledge is very solid, but there is no innovation ability, too dependent on books, and lack of critical thinking ability".

Therefore, although China's scientific research institutions can produce several times as many engineers and scientists as those of developed countries each year, this team is extremely weak in terms of 'technological' innovation ability, because it is impossible for a copier of 'scientific' ideas to have 'technological' originality and breakthrough, because there is no thinking training and ideological basis in this regard. It is no wonder that in some places, people can easily get stuck in our necks by using 'technology'.

Isn't it because we pay more attention to book science and less attention to technological innovation in our children's education?!

Outlook:

If "science" and "technology" are compared to the two supporting legs of a country's scientific and technological strength, then China's "technology" is still very weak! So, what should we do?

First of all, the weakness of China's "technology" leg is not congenital, let alone incurable! On the contrary, the four great inventions of the Chinese people, the invention of traditional Chinese medicine, just proves that we Chinese people are fully intelligent and have the ability to complement our 'technological' innovation weaknesses;

Secondly, when the Chinese nation is in danger, especially in the recent international scientific and technological competition, and even in the increasingly fierce scientific and technological war, the neck can only stimulate the intelligence and innovation ability of the Chinese people, and accelerate the improvement of our self-correction and self-innovation ability;

Finally, the path of rejuvenation of the Chinese nation is doomed to be bumpy, but there is no force that can prevent us from realizing our dream of becoming a powerful country. The ups and downs of the Chinese NMT people in the past two decades have also proved that the Chinese people can not only 'technological' innovation, but also step by step to the forefront of the world's scientific and technological innovation.

Yes, we may have been wrong in the past! Wrong method! Even in the wrong direction!

Isn't it true that human technology has made progress
in the process of constantly correcting mistakes?!

(Editor in charge: Xuefei Li)

Opportunities for Innovation

Changes of H⁺ flow rate in mesophyll cells of *Arabidopsis thaliana* under different frequencies of light

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Abstract: Different wavelengths of light have different biological effects, and have different effects on the morphological structure and photosynthesis of plants. Therefore, portable multi-spectral light processing instrument is developed by application, which can provide different wavelengths of light and changes in fixed frequency, and provide richer light conditions for experiments. In this paper, a portable multi-spectral light processing instrument is used to provide light of different wavelengths. *Arabidopsis thaliana* seedlings are used as materials. *Arabidopsis thaliana* mesophyll cells are irradiated with red, green, blue, white and purple light, and the real-time detection of H⁺ flow rate of mesophyll cells is performed using a non-invasive micro-measurement system. The results showed that under blue and white light, the absorption of H⁺ in mesophyll cells of *Arabidopsis thaliana* decreased, and under red and green light, the absorption of H⁺ was stronger, and under purple light, the absorption showed a trend of weakening, indicating that the flow rate of H⁺ in mesophyll cells was greatly affected by different color light. The study of this paper provides a basis for the study of photosynthesis and plant morphology and structure.

Keywords: *Arabidopsis*; H⁺; Current Speed; Non-damage micro-measurement technology; Portable multispectral light processing instrument

Light plays a very important role in plants. It plays a very important role in plant growth and development and in participating in plant photosynthesis. Therefore, this study is also to understand the impact of different frequencies of light on the physiological changes of plant leaves.

This experiment selected *Arabidopsis* as the test sample. *Arabidopsis* has a short growth cycle and is easy to grow. The construction and screening

Date of receipt: 2022-12-23; Received on: February 23, 2022

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process of its gene bank is simple and fast. Therefore, Arabidopsis is selected as the research object. Non-damage micro-measurement technology (NMT) is a technology for detecting living samples, which can detect the ion/molecular flow rate of living samples in real time, and ensure that the ion current changes of Arabidopsis leaf cells can be obtained at the instant of light treatment. For the selection of light frequency, a portable multi-spectral light processing instrument is selected, which can provide the change of light with fixed frequency and fixed time, providing accurate variable control for the experiment. In the experiment, hydrogen ion is selected as the detection index, and hydrogen ion participates in photosynthesis, which can detect the impact of light changes at different frequencies on photosynthesis.

1 Materials and methods

1.1 Plant materials

The seedlings of *Arabidopsis thaliana* were selected for the experiment. The seeds of *Arabidopsis thaliana*

were vernalized for 3 days. After vernalization, the seeds were sterilized with 5% sodium hypochlorite solution for 1 min, and then washed with sterile water for 5-6 times. The seeds were seeded in 1/2 MS medium. The seeds were irradiated for 16 hours and dark for 8 hours during the photoperiod; Light intensity is $300-400 \mu\text{mol cm}^{-2} \text{s}^{-1}$; The temperature under light conditions is 24°C and the relative humidity is 80%; The temperature under dark conditions is 20°C , and the relative humidity is 80%.

1.2 Different color light treatment

Portable multispectral light processing instrument (Xuyue (Beijing) Technology Co., Ltd.) (Figure 2), model PF-L-01, working voltage: 9V1A. The portable multi-spectral light processing instrument provides red, green, blue and purple monochrome light and white light with a total of five colors of light. The five colors of light change at a fixed frequency (60s/color light) in the order of red, green, blue, white and purple.

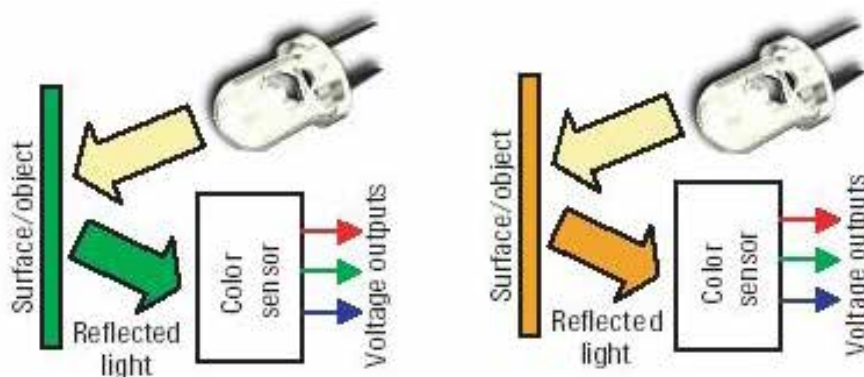


Figure 1: The color of reflected light depends on the color of surface reflection and absorption

A portable multispectral light processing instrument was used to carry out real-time cyclic light processing on Arabidopsis mesophyll cells.



Figure 2: Portable multispectral light processing instrument

1.3 Measurement of H⁺ flow rate of Arabidopsis mesophyll cells

Utilize non-invasive micro-test technology (NMT), and use the seventh generation product NMT Physiolyzer[®] (NMT in vivo physiological detector, Younger, Youngerusa. com) to test the flow rate data. Take the leaves of Arabidopsis to be tested, determine a small area of the leaves to be tested, tear off the lower epidermis, and expose the mesophyll tissue. Prepare a petri dish, add the test solution (0.1 mM KCl, 0.1 mM CaCl₂, 0.1 mM MgCl₂, 0.5 mM NaCl, 0.3 mM MES, 0.2 mM Na₂SO₄, pH 6.0), put the prepared leaf tissue with the lower surface downward, let it float on the test solution, the lower surface fully contact the test solution, and leave it standing for 15 minutes, then discard the test solution. Take out the leaf tissue carefully, fix the leaf tissue in the petri dish, add 5ml of fresh test solution, and load

the sample for testing. Find the target detection area under the microscope, and place the H⁺ flow sensor at a distance of about 5 from the mesophyll cell surface μ At m, the test is started (Figure. 3). Each sample is tested for 5-10 minutes, during which the portable multispectral light processing instrument is used to process different colors of light, and each group is tested for 5 replicates. The H⁺ flow rate data is directly read through imFluxes V2.0 software (YoungerUSA LLC, Amherst, MA 01002, USA). The flow rate unit is mol • cm⁻² • s⁻¹. Positive value represents efflux and negative value represents absorption.

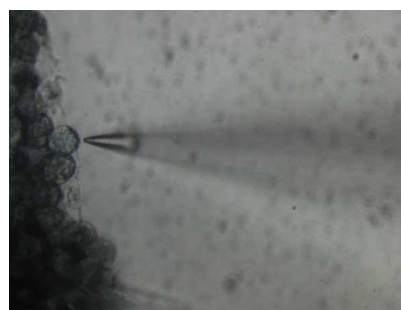


Figure 3: Portable multispectral light processing instrument

1.4 Data analysis

The H⁺ flow rate data uses the flow rate cloud (IF100, Weishui (Beijing) Technology Co., Ltd, <http://nmtia.org.cn/imfluxes>)Software processing and analysis.

2 Results and discussion

2.1 Effect of different color light on H⁺ flow rate of Arabidopsis mesophyll cells

The experimental results show (Figure . 4) that under the fixed frequency (60s/color light) in the order of red, green, blue, white and purple, under the blue and white light, the absorption trend of H^+ in the mesophyll cells of *Arabidopsis thaliana* decreases, under the red and green light, the absorption of H^+ increases, and under the purple light, the absorption trend decreases. In the second cycle of the same wavelength and frequency, this phenomenon occurs.

Note: The positive value of the flow rate represents efflux, and the negative value represents absorption. The data obtained in the experimental results are negative values, both of which are absorption trends.

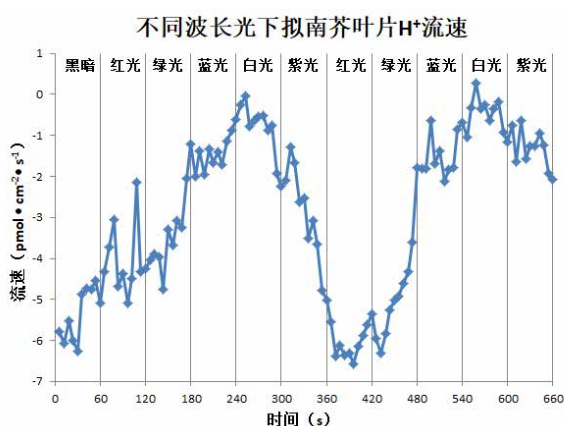


Figure . 4 H^+ flow rate of *Arabidopsis* mesophyll cells under different wavelength light

2.2 Discussion

The different wavelengths of light discussed in this paper are of concern to researchers. In this paper, when plant leaves are exposed to different wavelengths of light, H^+ absorption is closely related to the wavelength of light. However, only the fixed change of wavelength can not meet the needs of scientific researchers. The adjustment of optical frequency, the order of different wavelengths, and more wavelengths of light are all the needs of scientific researchers for experiments.

The portable multispectral light processing instrument has not been made into an adjustable product for the light intensity. The light intensity is one of the indispensable parameters in the experiment. The portable multispectral light processing instrument has also been improved on the function of light intensity. It is expected that the latest version of the product can help scientific researchers solve more problems.

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(Editor in charge: Xuefei Li)

Effect of antennal temperature change on Ca²⁺ signal of Indian corn borer

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Abstract: In order to solve the difference of experimental results caused by environmental differences, this paper uses a newly developed experimental environment monitor to detect the environmental parameters of living samples in real time during the experiment to ensure the recording of environmental parameters in the experiment. This paper uses the experimental environment monitor to detect the Ca²⁺ flow rate of insect antennae with NMT while detecting the physiological environment of insects. Under the environment of temperature 25 °C, humidity 29%, pressure 995hPa, and altitude 145m, the Ca²⁺ flow rate is detected as an absorption trend, while under the environment of temperature 15 °C, humidity 54%, pressure 994hPa, and altitude 153m, The absorption of calcium ions by insect antennae is significantly higher than that of the environment at 25 °C. This paper provides a research basis for studying the effects of changes in different physiological environment parameters on the physiological functions of organisms.

Key words: Non-invasive Micro-test Technology; Environmental parameters; Temperature; Humidity; Pressure; Altitude; Experimental environment

Indian corn borer is a relatively common moth in China, belonging to the insect class Lepidoptera mothidae. It has not been found in Tibet, and is distributed in other provinces, cities and autonomous regions in China. It harms all kinds of grains and processed products, beans, oilseeds, peanuts, all kinds of dried fruits, dried vegetables, milk powder, preserved fruits, traditional Chinese medicine, tobacco, etc. Indian grain borer can grow and develop in different environments. This experiment is also to understand the physiological changes of Indian grain borer under different temperature environments, and then understand the impact of environment on insects. Insects can react quickly to the change of

environmental temperature. In the experiment, we need to select the technology that can not only ensure the active state of insects, but also detect the corresponding indicators. Therefore, we use the Non-invasive Micro-test Technology (NMT), which can quickly detect the molecular ion flow rate of living samples, and ensure that insects can

Date of receipt: 2022-12-23; Received on: February 23, 2022

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detect the corresponding indicators under the active state. In order to capture and record the changes of the environment faster and more accurately, the experimental environment monitor (PEP) is selected to record the changes of the environment in real time and output the real-time changes of the environment.

In the selection of indicators, calcium ions participate in the synthesis and release of synaptic neurotransmitters, the synthesis and secretion of protein hormones, metabolism and the activation and release of intracellular and extracellular enzymes, and calcium ions participate in the regulation of pain information in neurons. For the selection of signal indicators of insect antennae, calcium ions are selected as detection indicators.

The following experiment is based on the design of the combination of living body and environment, and provides a method for the study of the impact of environment on insects.

1 Materials and methods

1.1 Materials

The Lepidoptera boreidae insects (Figure. 1) were selected for the experiment. The samples were collected from newly hatched insects in the field, and the photoperiod was 16 hours of light and 8 hours of darkness; Light intensity is $300-400 \mu \text{mol cm}^{-2} \text{s}^{-1}$; The temperature under light conditions is 24°C and the relative humidity is 80%; The experiment was

carried out after two days in an environment with a temperature of 20°C and a relative humidity of 80% in the dark.



Figure 1: Lepidoptera boreidae insect samples

1.2 Experimental environment monitor

The experimental environment monitor (PEP) (Xuyue (Beijing) Technology Co., Ltd.) (Figure 2) is PEP-1.0. PEP can detect and display the temperature, humidity, pressure and altitude values of the surrounding environment of living samples, and can display the broken line chart of data and time on the web page and download the CSV file.

In the experiment, PEP is used to detect various parameters in the environment to provide more accurate parameter basis for the experiment.



Figure 2: Experimental environment monitor

1.3 Determination of Ca²⁺ flow rate of insect antennae

Utilize non-invasive micro-test technology (NMT), and use the seventh generation product NMT Physiolyzer[®] (NMT in vivo physiological detector, Younger, Youngerusa. com) to test the flow rate data. Prepare a petri dish, add the test solution (0.1 mM KCl, 0.1 mM CaCl₂, 0.1 mM MgCl₂, 0.5 mM NaCl, 0.3 mM MES, 0.2 mM Na₂SO₄, pH 6.0), immerse the insect in the test solution to expose the area to be measured by the head antenna, and leave it standing for 15min, discard the test solution, add 5ml of fresh test solution, and load the sample for testing. Find the target detection area under the microscope, and place the Ca²⁺ velocity sensor about 5 from the end surface of the insect antenna μ At m, the test is started, and each sample is tested for 3 minutes. During the treatment under normal environment and low temperature environment, 5 replicates were detected in each group. The Ca²⁺ flow rate data is directly read through imFluxes V2.0 software (YoungerUSA LLC, Amherst, MA 01002, USA). The flow rate unit is mol • cm⁻² • s⁻¹. Positive value represents efflux and negative value represents absorption.

1.4 Data analysis

Ca²⁺ flow rate data is based on flow rate cloud (IF100, Weishui (Beijing) Technology Co., Ltd, <http://nmtia.org.cn/imfluxes>) Software processing and analysis.

2 Results and discussion

2.1 Experimental results

Using NMT to detect the Ca²⁺ flow rate of insect antennae (Fig. 3), under the environment of temperature 25 °C, humidity 29%, pressure 995hPa, and altitude 145m, it is detected that the calcium ion flow rate is an absorption trend, while under the environment of temperature 15 °C, humidity 54%, pressure 994hPa, and altitude 153m, the absorption of calcium ion by insect antennae is significantly greater than that under the environment of temperature 25 °C.

Note to Figure 3: A is a low temperature environmental parameter; B is the environmental parameter under normal temperature; C is the Ca²⁺ flow rate of insect antennae in different environments, where the positive value of the flow rate represents efflux and the negative value represents absorption.

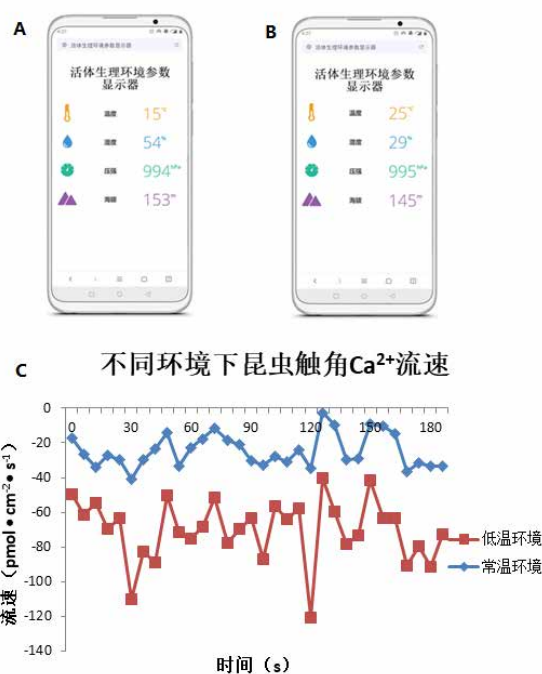


Figure 3: Ca²⁺ flow rate of insect antennae in different environments

2.2 Discussion

The parameters of the external environment have always been the focus of researchers, but the comprehensive measurement products of the external environmental parameters are relatively few. The parameters that the experimental environmental monitor (PEP) can provide are still not comprehensive. In the face of other environmental parameters such as longitude and latitude, atmospheric oxygen concentration, carbon dioxide concentration, etc. are still in the process of development and improvement, and other parameters and functions also need scientists to provide innovative ideas, Products that jointly improve environmental parameters.

This article uses the latest generation of PEP products, which are not perfect in real-time display of environmental parameters and data output. The latest generation of PEP products has added this function to ensure that the real-time environmental parameter values are corresponding to the sample data, so that the environmental parameters of the data are more accurate. At the same time, PEP products are also trying to output on the mobile phone. If there are functions that need to be improved in the experiment, we still need a large number of scientific researchers to provide the demand.

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(Editor in charge: Xuefei Li)

New to NMT

Analysis of Multi-sensor Detection Function in Non-damage Micro-measurement System

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Recently, I am studying an article ^[1] by Professor Wang Guangce of the Institute of Oceanography, Chinese Academy of Sciences. This article uses the non-invasive micro-measurement system (hereinafter referred to as NMT system) to detect the H^+ and O_2 flow rates of *Zostera marina* (eelgrass). Interestingly, in this study, the flow rates of H^+ and O_2 were obtained by simultaneous detection of the same site of the sample using hydrogen and oxygen sensors, which is very rare and innovative. However, today we will not talk about the scientific significance of dual-sensor detection. Let's talk about what is multi-sensor, as well as the questions and misunderstandings in the process of selecting multi-sensor functions.

I. Concept

Multi-sensor detection, also known as multi-channel detection or multi-electrode detection, refers to the function of NMT system to detect several or more ionic molecules simultaneously for the same sample. It usually includes three-sensor detection, four-sensor detection, multi-sensor detection, etc.

II. Classification

The multi-sensor detection of NMT system is mainly divided into two categories:

1. Use multiple sensors to simultaneously detect different parts of the same sample or different sites of the same part.
2. Using multiple sensors to detect the same site of the same sample at the same time, Professor Yin Liping of Capital Normal University elaborated this dual-sensor detection method in his article ^[2].

The common feature of the two types of dual-sensor detection is that they both use multiple sensors to detect the same sample at the same time. The difference is that the latter can detect the same site, while the former cannot achieve it. Therefore, the latter is also known as multi-sensor detection at the same site. The dual-sensor detection of the NMT system used in the above article belongs to this category.

Date of receipt: 2022-12-23; Received on: February 23, 2022

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III. Function

In order to realize the multi-sensor detection function, the NMT system must have multiple channels. However, it is impossible to realize the multi-sensor detection function by only having 3 channels, 8 channels or multiple channels. To achieve this function, the problem of multi-sensor array and multi-signal acquisition must be solved. It is understood that at present, relevant technical problems have been solved, and there are patents related to multi-channel or multi-sensor detection [3].

In order to realize the multi-sensor detection function at the same location, the NMT system should not only have multiple channels, but also solve the problem of multiple sensor arrangement and multiple signal acquisition, but also break through a technical problem, that is, how to locate multiple sensors at the same location. According to the certification standards [4] issued by the Zhongguancun NMT Industry Alliance and the relevant funds, the products on the market can only achieve the same-point dual-sensor detection, and the same-point multi-sensor detection is still in further development.

IV. Function selection

Teachers should pay attention to the following aspects when selecting the multi-sensor detection function of NMT system:

1. When selecting the multi-sensor detection function, be sure to check with the merchant and indicate in the function parameters the number of sensors to be detected at the same time, not the number of channels. For example, three sensors are required to detect at the same time instead of three channels.
2. If you want to repeat Professor Wang Guangce's experiment, you must select the function of "same-point dual-sensor detection". Multi-sensor detection cannot achieve this function.
3. Select products certified by Zhongguancun NMT Industry Alliance.

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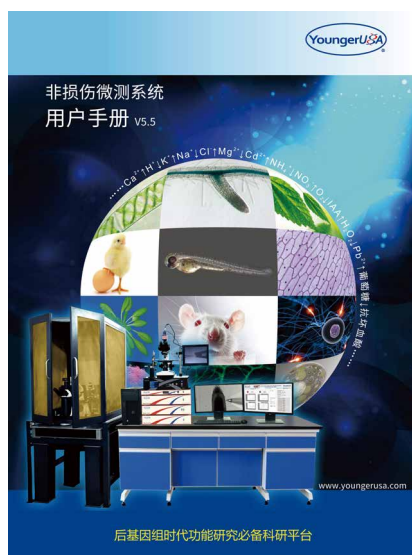
(Editor in charge: Xuefei Li)

Equipment maintenance - NMT system maintenance and repair tips

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The best way to prolong the service life of the system is to operate in strict accordance with the process and good use habits!



I think everyone should be familiar with this user manual of the non-invasive micro-measurement system. As long as the user who purchases the system, regardless of the model, will attach a copy with the system (the legend is the latest version of the manual).

But how many people have really read this manual carefully from beginning to end? I think it is rare. After the system operation training, most people have mastered the basic test operation process, and may not read this manual again. They think it should be useless.

In fact, this is a big mistake! This manual is the crystallization of Xuyue's technology and experience accumulated over the past 20 years. In addition to the basic operation of the test, some precautions in the test and solutions to problems encountered are recorded in detail. Especially in the process of system operation, there are some

Date of receipt: 2022-12-23; Received on: December 33, 2022

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operation procedures and use specifications that may not be paid much attention to at ordinary times, but in the long run, it may cause great damage to the system hardware, shorten the service life of accessories, cause system failure and other problems.

Therefore, it is a necessary step to read the user manual carefully before operating the system. Please take it seriously. The operation shall be carried out in strict accordance with the process specifications in the manual, and good use habits shall be formed, which can not only prolong the service life of the system, but also greatly help your own experiment. At the same time, new users should choose the formal training class of Xuyue Company when learning the system operation to ensure the training effect and quality.

(Editor in charge: Xuefei Li)

Daily use and maintenance of data acquisition system

1. Pay attention to dust prevention

After each use, cover the dust cover attached to the system and close the laboratory doors and windows.

2. Regular cleaning

Tools required: a cross screwdriver (need to be purchased separately)

Specific steps:

- 1) Turn off the power, remove all cables of the data acquisition system, and place it on a spacious desktop.
- 2) Unscrew the six screws marked in the figure below with a cross screwdriver.

Remove the rear panel after all 6 screws are unscrewed.

Note: In order to avoid the loss of screws, it is better to put them in small bags or other containers.



Rear panel fixing screws of data acquisition system

- 3) Unscrew the four screws marked in the figure below.



Data acquisition system side panel fixing screws

- 4) Remove the side panel, as shown below

Note: Do not use brute force, push out in the direction of the arrow.



Remove the side panel

- 5) Use the air compression tank or ear ball (need to be purchased separately) to clean the inside of the case, especially the fan and dusty places. After completion, follow the above steps to install the chassis panel back, connect the power cord, monitor, mouse, keyboard, and start up and run. If the machine is started normally, install the remaining connecting wires back to the original position.

Next notice!

Common sense of daily use and maintenance of sensor
preparation system!

(Editor in charge: Xuefei Li)

Experimental system: detection of hydrogen ion secretion rate of mouse gastric mucosa

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I. Purpose of the experiment

To understand the detection method of hydrogen ion in mouse gastric acid secreting glands.

II. Materials, appliances, instruments and drugs

Test sample: mouse gastric mucosa

Detection instrument: non-damage micro-measurement system (scientific research platform) or non-damage micro-measurement system (research and development platform)

Consumables: NMT dedicated solid H⁺flow sensor

Required drugs: 141.8 mmol · L⁻¹ NaCl, 5.9 mmol · L⁻¹ KCl, 1.2 mmol · L⁻¹ MgCl₂, 10 mmol · L⁻¹ HEPES acid, 5.6 mmol · L⁻¹ Tris, 2.56 mmol · L⁻¹ CaCl₂, 2 g glucose, pH 7.4 100 mmol · L⁻¹ histamine, 300 mmol · L⁻¹ cimetidine.

Tools: stereomicroscope, silica gel plate, culture dish, CO₂, pipette gun, several gun heads.

III. Principle

The gastric mucosa is mainly composed of parietal cells that secrete gastric acid, main cells that secrete pepsinogen and mucous cells that secrete mucus. These cells coordinate with each other to participate in the digestive process in the stomach. Gastric acid (HCl) is derived from parietal cells and is produced through the synergy of multiple transporters and ion channels in the apical and basal membranes of parietal cells. Under stimulation, H⁺/K⁺-ATPase, Cl⁻ secretion pathway, K⁺ circulation pathway and Cl⁻ absorption pathway of basal membrane of parietal cells participate in the secretion of gastric acid. Histamine can stimulate the secretion of gastric acid. By binding with specific receptors on the parietal cell membrane, it can increase the level of cAMP in the cell, activate the ion channels and transporters on the cell membrane, and increase the secretion of HCl.

IV. Method steps

1. Preparation of isolated gastric mucosa: mice were euthanized by inhaling carbon dioxide. Take out the stomach, keep the stomach body, cut the stomach body along the great curvature of the stomach, fix it on the silica gel plate after cleaning, and separate the mucosa layer under the posture microscope.
2. Put the gastric mucosa into a culture dish containing 5mL of test solution and fix it for 30min.
3. After replacing the test solution, put it into the NMT system to detect the flow rate of H^+ in the facial mask of the gastric mucosa.
4. Instantly add the final concentration of 100mM histamine, and continue to detect the H^+ flow rate for 10min.
5. Then add cimetidine with a final concentration of 300mM and continue to carry out H^+ flow rate detection.

V. Experimental report

According to the experimental results, explain the physiological phenomenon and principle of H^+ change after adding histamine and cimetidine.

VI. Expected results

After histamine treatment, the secretion of H^+ increased; After further addition of cimetidine, H^+ secretion gradually decreased.

VII. Thinking questions

1. Why can NMT quickly detect the gastric mucosal H^+ flow rate?

Answer: NMT is a technology with ultra-high sensitivity, non-contact method, based on the flow rate, to detect the molecular concentration and gradient of external ions of materials, which can quickly detect the changes of sample molecular ions.

2. Can NMT detect the hydrogen ion flow rate of cultured parietal cells, and is there any difference between NMT and the detection of acutely isolated gastric mucosa tissue?

Answer: It can detect cultured parietal cells. The normal growth of gastric mucosal parietal cells and the cultured parietal cells will differ in activity. The data reflected by the normal growth of gastric mucosal parietal cells will be closer to the real live data.

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(Editor in charge: Xuefei Li)

NMT Communications
March 2023 Volume 1 Issue 2

Publisher: Non-invasive Micro-test Technology International Alliance
Publisher Address: 19 Research Drive, Suite 6 Amherst, MA 01002

Organizer: Zhongguancun Xuyue Non-invasive Micro-test Technology Industrial Alliance
Editor-in-Chief: Xu Yue and Liu Yunqi
Honorary Editors: Kuang Tingyun, Sergey Shabala, Marshall Porterfield
Address of editorial office: 19 Research Drive, Suite 6 Amherst, MA 01002

Executive Editor: Li Xuefei
Editors: Zhang Zengkai, Gao Hongjian, Li Lei, Ye Bin, Ju Xiaoyu, Ma Yue, Guo Weiwei, Liu Peiqi
Art Editors: Liu Zhaoyi, Wang Yang

International Standard Serial Number (Online): ISSN2834-5363
International Standard Serial Number (Print): ISSN2834-5355
Submission email: yanhan@nmtia.org.cn
Tel: 010-8262 4800

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December 30, 2022