

Efficiency improvement starts from standardization
Introduction to the Standard Operating Procedure for Classification of NMT
Experiments in Plant Research (SOP)

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Abstract: The aim of this study is to explore the application of Non-invasive Micro-test Technology (NMT) in plant physiology research. With the help of the International NMT Alliance and the Zhongguancun NMT Industry Alliance, and under the direct guidance of the International NMT Standardization Committee (NISC), a standard experimental protocol is provided for first-time researchers who are exposed to this technology. By organizing and summarizing the standard operating procedures (SOPs) for different experimental needs in different research directions, including salt stress, heavy metal stress, plant nutrition, reproductive development, guard cells, proton pumps, and calcium signaling, based on published literature results, the success rate and efficiency of NMT experiments are improved, and the progress of plant physiology research is promoted.

1. Introduction

Non-invasive Micro-test Technology, as an important biological research tool, provides researchers with a new means of in vivo and real-time physiological research on the microenvironment. In order to facilitate researchers to quickly get started and obtain objective and authentic experimental data, with the help of the International NMT Alliance and the Zhongguancun NMT Industry Alliance, and under the direct guidance of the International NMT Standardization Committee (NISC), the experimental standard operating procedures (SOPs) were developed based on the research directions, materials, and indicators that are currently widely used in NMT.

2. Classification and description:

Salt stress research: This study developed a series of standard operating procedures (SOPs) based on different research directions. Research direction on salt stress: Salt stress has a serious impact on plant growth and development. We have developed an SOP that covers parameters related to salt stress, including Na⁺ rate, K⁺ rate, and H⁺-ATPase activity in root and mesophyll rows.

Research on heavy metal stress: The toxic effects of heavy metal pollution on plants have received much attention. We provide a standard operating procedure for measuring the rate of heavy metal uptake by roots and the activity of H⁺-ATPase, which helps to evaluate the plant's ability to cope with heavy metal stress.

Nutrient element research: The absorption and transportation of nutrients by plants are

crucial for their growth. We have developed SOPs for the rate of ammonium nitrate uptake by roots and the activity of H⁺-ATPase, providing guidance for studying the nutrient absorption mechanism of plants.

Reproductive growth and development research: Reproductive development is a critical stage in the plant life cycle. We have provided a detailed introduction to the SOP for pollen tube and stigma NMT experiments, providing technical support for studying plant reproductive processes.

Guard cell research: Guard cells play an important role in plants' response to environmental changes. We provide a standard operating procedure for measuring the ion/molecule transmembrane transport rate during stomatal opening and closing processes, providing methodological support for exploring plant defense mechanisms.

Proton pump/H⁺-ATPase research: Proton pumps play an important role in maintaining intracellular and extracellular ion balance. We have developed an SOP for the H⁺rate/H⁺-ATPase activity in the root system, providing experimental guidance for studying the regulatory mechanism of proton pumps.

Calcium signaling research: Calcium signaling plays a crucial role in plants' response to external stimuli. We have developed an SOP for the Ca²⁺transport rate in roots and leaves, which helps to further explore the calcium signaling mechanism in plants.

3. Conclusion:

The classification standard process for NMT experiments in different research directions introduced in this article has been published on the official website of the Zhongguancun NMT Industry Alliance (<http://nmtia.org.cn/>) Showcasing, from experimental preparation to operational steps, provides standardized operational guidelines for beginners. The development of these standard operating procedures not only accelerates experimental progress and improves experimental efficiency, but also helps to promote the in-depth development of plant research. Through the standard operating procedures provided in this article, researchers can better utilize NMT technology to deeply study the physiological and ecological characteristics of plants, and contribute to the progress of agriculture and environmental protection.

Reference:

- 1.Chen G, Xuan W, Zhao P, Yao X, Peng C, Tian Y, Ye J, Wang B, He J, Chi W, Yu J, Ge Y, Li J, Dai Z, Xu D, Wang C, Wan J. OsTUB1 confers salt insensitivity by interacting with Kinesin13A to stabilize microtubules and ion transporters in rice. *New Phytol.* 2022 Sep;235(5):1836-1852.
- 2.Li L, Mao D, Sun L, Wang R, Tan L, Zhu Y, Huang H, Peng C, Zhao Y, Wang J, Huang D, Chen C. CF1 reduces grain-Cd levels in rice (*Oryza sativa*). *Plant J.* 2022 Mar 16.
- 3..Liu B, Feng C, Fang X, Ma Z, Xiao C, Zhang S, Liu Z, Sun D, Shi H, Ding X, Qiu C, Li J, Luan S, Li L, He K. The anion channel SLAH3 interacts with potassium channels to regulate

nitrogen-potassium homeostasis and the membrane potential in Arabidopsis. *Plant Cell*. 2023 Jan 19:koad014

4 .Meng JG, Liang L, Jia PF, Wang YC, Li HJ, Yang WC. Integration of ovular signals and exocytosis of a Ca^{2+} channel by MLOs in pollen tube guidance. *Nat Plants*. 2020 Feb;6(2):143-153. doi: 10.1038/s41477-020-0599-1. Epub 2020 Feb 13. PMID: 32055051.

5. Pei D, Hua D, Deng J, Wang Z, Song C, Wang Y, Wang Y, Qi J, Kollist H, Yang S, Guo Y, Gong Z. Phosphorylation of the plasma membrane H^{+} -ATPase AHA2 by BAK1 is required for ABA-induced stomatal closure in Arabidopsis. *Plant Cell*. 2022 Apr 11:koac106.

6. Fu L, Wu D, Zhang X, Xu Y, Kuang L, Cai S, Zhang G, Shen Q. Vacuolar H^{+} -pyrophosphatase HVP10 enhances salt tolerance via promoting Na^{+} translocation into root vacuoles. *Plant Physiol*. 2022 Feb 4;188(2):1248-1263. doi: 10.1093/plphys/kiab538. PMID: 34791461; PMCID: PMC8825340.

7. Li J, Shen L, Han X, He G, Fan W, Li Y, Yang S, Zhang Z, Yang Y, Jin W, Wang Y, Zhang W, Guo Y. Phosphatidic acid-regulated SOS2 controls sodium and potassium homeostasis in Arabidopsis under salt stress. *EMBO J*. 2023 Feb 22:e112401. doi: 10.15252/emboj.2022112401. Epub ahead of print. PMID: 36811145.

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Fujian A&F University Continues to Open Testing Services to the Outside World for Non-invasive Micro-test Equipment

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In 2023, a Non-invasive Micro-test Technology engineer from Xuyue (Beijing) Sci.&Tech. Co., Ltd. came to Fujian A&F University for routine maintenance and on-site guidance of Non-invasive Micro-test Technology equipment.

Fujian A&F University is located at the gateway of the Maritime Silk Road and one of the first coastal port cities to open up to the outside world - Fuzhou. It is a provincial-level key university with the advantages and characteristics of agriculture and forestry disciplines, and coordinated development of multiple disciplines such as science, engineering, economics, management, literature, law, and art. It is a university jointly built by the Ministry of Agriculture and Rural Affairs, the National Forestry and Grassland Administration, and the Fujian Provincial Government, and a first-class university in Fujian Province



The NMT equipment of the school was purchased in 2015 and is currently located in the Key Laboratory of Agricultural Ecological Process and Safety Monitoring in Fujian Province. Sample testing is open to the public, and there is a dedicated person responsible for equipment operation



During this engineer's visit, in addition to routine equipment maintenance, three new equipment users and operators in the laboratory were provided with operational guidance to facilitate the continuation of external testing services in the future.

At present, the colleges in Fujian Agricultural and Forestry University that have used NMT technology include the College of Life Sciences, the College of Horticulture, the College of Plant Protection, the College of Agriculture, and the Metabolomics Research Center of the Joint Research Institute of the Taiwan Strait. More than 20 NMT literature have been published, covering application fields such as plant stress, heavy metal stress, plant nutrition, and plant microbial interaction.

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Preliminary Study
On the Qualification Certification System of the International NMT Alliance
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International NMT Alliance

Abstract: The "International NMT Alliance Qualification Certification System" is an important component of NMT's global scientific research and innovation, with the goal of standardizing the design, production, sales, and services related to NMT technology to ensure its efficiency and sustainable development. The content includes: innovation center qualification certification, equipment manufacturer qualification certification, standardization center qualification certification, testing service engineer qualification certification, training engineer qualification certification, etc., and is aimed at global technology practitioners. After obtaining the corresponding qualifications, one can meet the corresponding entry threshold or provide corresponding services.

Keywords: Non-invasive Micro-test Technology, NMT, qualification, certification

1. Certification system

The "International NMT Alliance Qualification Certification System" is an important component of NMT's global scientific research and innovation, with the goal of standardizing the design, production, sales, and services related to NMT technology to ensure its efficiency and sustainable development. The content includes: innovation center qualification certification, equipment manufacturer qualification certification, standardization center qualification certification, testing service engineer qualification certification, training engineer qualification certification, etc., and is aimed at global technology practitioners. After obtaining the corresponding qualifications, one can meet the corresponding entry threshold or provide corresponding services. Each qualification is divided into three levels: initial, intermediate, and advanced. It is necessary to apply for certification to the International NMT Alliance step by step. The International NMT Alliance will evaluate or assess the applicant or applicant unit according to the corresponding certification standards. After passing the evaluation, the applicant will obtain the corresponding certification qualification.

2. Certification project

- A. Innovation Center Qualification Certification: Relying on the innovation platform of NMT, possessing the qualification to undertake research and application innovation needs, and provide various NMT related services;
- B. Standardization Center Qualification Certification: Promote the transformation of NMT related achievements for the purpose of publishing standards, and have the qualification to provide services for the formulation of NMT related groups and industry standard requirements;

- C. Equipment manufacturer qualification certification: Evaluate the comprehensive performance of NMT equipment produced, as well as successful cases in various aspects such as software and hardware, consumables, manufacturer after-sales service, research and development capabilities, market data, etc., to provide guidance and guidance for original and integrated innovation in scientific research, procurement of NMT equipment/consumables/services, and fair and healthy competition among relevant production and service enterprises.
- D. Qualification certification for testing service engineers: To provide technical service engineers at all levels with corresponding capabilities for global NMT experimental needs, ensuring high-quality and high standard NMT data output;
- E. Training Engineer Qualification Certification: To meet the global demand for NMT equipment usage, we provide training engineers at all levels with corresponding capabilities to ensure the use of NMT equipment by various institutions worldwide, and to produce high-quality and high standard NMT data.

NMT, as an emerging technology with broad application prospects, has produced rich research results in global animal, plant, medical, ecological environment, microbiology, materials and other scientific research fields, and has derived more demands for NMT technology applications/experiments/services. The "International NMT Alliance Qualification Certification System" ensures that global researchers obtain reliable and effective NMT experimental data, and improves the level of scientific research in various fields worldwide, requiring everyone's active participation

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