

Application Status and Prospects of Rapid Test Kits for Crop Diseases

Qiaolian Pang, Daoyu Xu, Hongli Li, Shihui Wang*

Shandong Zhongxinkenong Life Technology Co., Ltd., Jinan, Shandong, 250100, China

*Corresponding author: Shihui Wang, sdzxkn@sdzxkn.com

Copyright: 2024 Author(s). This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY-NC 4.0), permitting distribution and reproduction in any medium, provided the original author and source are credited, and explicitly prohibiting its use for commercial purposes.

Abstract: Rapid test kits for crop diseases are agricultural tools for early monitoring of crop diseases. Their application can reduce the use of pesticide, mitigate environmental pollution, and promote green and sustainable agricultural development. This paper reviews the research progress, categories, and applications of rapid detection kits for crop diseases, as well as existing challenges and solutions. Additionally, it provides insights into the future prospects of portable rapid test kits for crop diseases, aiming to offer valuable references for their development.

Keywords: Crop Diseases; Rapid Test Kits; Research Progress; Prospects

Published: Apr 15, 2024

DOI: https://doi.org/10.62177/apemr.v1i2.268

1.Introduction

Most of the food that humans rely on for survival comes directly or indirectly from the roots, stems, leaves, flowers, seeds, and fruits of crops. The safety of various organs of crops affects human life and health. In recent years, green, low-carbon, and sustainable development has become the trend of global economic development. The pursuit of green and sustainable development in agriculture contributes to enhanced food safety. According to the speculation of the Food and Agriculture Organization of the United Nations, plant diseases and pests cause an annual loss of 20% to 40% of the world's crop production, affecting the main crops in countries and regions on all continents of the world ^[1-2]. Crop disease rapid detection kits can diagnose crop diseases at an early stage, enabling farmers to take timely and effective prevention and control measures based on the detection results. This can effectively avoid yield and economic losses. It is an important measure to help farmers achieve a bumper harvest, reduce the use of pesticides, and promote the green and sustainable development of agriculture.

In recent years, portable crop disease rapid test kits have been successively launched in the agricultural markets of countries such as the United States, the United Kingdom, and South Korea. At the end of 2021, the dot-ELISA rapid detection kit developed by Zhou Xueping's team for one major crop virus (Southern Rice Black-Streaked Dwarf Virus) and three quarantine crop viruses (Maize Chlorotic Mottle Virus, Tomato Brown Rugose Fruit Virus, and Cucumber Green Mottle Mosaic Virus) came out, ending the situation in which China could only rely on imports for crop disease rapid test kits ^[3]. In order to ensure national food security and the quality and safety of agricultural products, protect the ecological environment, and promote the sustainable development of agriculture, the State Council announced the Regulations on the Prevention and Control of Crop Diseases and Pests on April 3, 2020. These regulations established a monitoring system for crop diseases

and pests, emphasizing that the prevention and control of crop diseases and pests should follow the principle of prevention first and comprehensive prevention and control, and carry out green prevention and control supported by science and technology ^[4]. Crop disease rapid test kits can help agricultural producers and other relevant units and individuals do a good job in the early diagnosis, monitoring, and early warning of crop diseases. Their application can not only provide targeted guidance for farmers to use pesticides correctly, reduce the amount of pesticides used, and reduce the waste and pollution of pesticides, but also reduce the planting cost of crops and ensure the yield. It is one of the important measures to achieve green agriculture. However, currently in China, crop disease rapid test kits are still mainly in the stages of experimental research and government inspection and quarantine applications, and they are still very rare in agricultural input retail stores. Based on market research and online searches, this article predicts and analyzes the research progress, applications, existing problems, and prospects of crop disease rapid test kits, hoping to provide references for the research and development in this field.

2. The Concept and Research Progress of Crop Disease Rapid Test Kits

2.1 The Concept of Crop Disease Rapid Test Kits

The crop disease rapid test kit is a kit that quickly determines the presence of certain pathogens (fungi, bacteria, nematodes and viruses) in crops.

The well-known COVID-19 detection kit works by dipping a sample from the human nasal cavity into the reagent in the kit, shaking it well, and then dropping the reagent into the corresponding area of the kit to observe whether a horizontal line appears in the detection line to determine whether the person is infected with the COVID-19 virus. For a crop disease rapidtest kit, samples are taken from areas such as the soil near the roots of crops, the root tissues of crops, or the leaf tissues of crops. After sampling, the sample is put into the reagent in the kit, shaken well, and then the reagent is dropped into the designated area of the kit. Within a few minutes, by observing whether a horizontal line appears in the detection area of the kit, it can be determined whether the crop is infected with a certain plant pathogen.

2.2 Research Progress of Crop Disease Rapid Test Kits

The research on crop disease rapid test kits began in the early 1980s, originally stemming from the development of monoclonal antibodies against tobacco mosaic virus by German scientists^[5]. In 1985, Diaco et al. developed an Elisa kit for detecting soybean mosaic virus in soybean seeds^[6]. Richard et al's^[7] application for a crop disease diagnostic kit was granted a patent in the US on 10 September 1991. On June 15, 1993, another patent application by Richard's^[8] team, entitled "Method and a kit for diagnosing plant diseases," was also granted in US. On September 4, 2020, a kit for the simultaneous diagnosis of Fusarium oxysporum and Ralstonia solanacearum by a semi-quantitative lateral flow immunoassay applied for by Kim^[9] et al. was granted a patent in South Korea. The crop disease rapid test kit developed by them has been marketed in Korea, Colombia, Cambodia, Vietnam and other countries. On March 19, 2021, Razo et al.^[10] developed a new rapid detection test strip for diagnosing Erwinia amylovora (also known as fire blight) by a lateral flow immunoassay.

Although the research on crop disease detection technology in China started relatively late, it has developed rapidly and achieved great progress in recent years. In 1995, Wei ^[11] reported the technology of nucleic acid spot hybridization for detecting GFV and PVY using biotin-labeled double-stranded cDNA probes synthesized by PCR technology and a chemiluminescence system. In 1997, Zhang et al.^[12] developed a colloidal gold rapid test kit for Huanglongbing bacteria. In 2001, Li et al. ^[13] developed an alkaline phosphatase enzyme-linked diagnostic kit for Tobacco ringspot virus and Potato virus X. This was the first kit developed in China using alkaline phosphatase labeling and the DAS-ELISA method. In 2006, Wang Lin et al. applied for patents on gene chips, nucleotide sequences and kits for detecting viruses in solanaceae crops ^[14] and leguminous crops ^[15], respectively, and were granted in 2010. However, the above two patents are mainly used for daily quarantine work on plant-derived food, and are not applicable to on-site detection of viruses in crops grown in the field. Wei et al. ^[16] prepared a colloidal gold chromatography test strip that can quickly and sensitively identify the BBTV virus in banana plants. This test strip has the same detection limit as fluorescent PCR. Tian ^[17] prepared a colloidal gold immunochromatography test strip for detecting Potato virus V. Ruan et al. ^[18] developed double and triple colloidal gold test strips that can simultaneously detect Tobacco mosaic virus (TMV), Cucumber mosaic virus (CMV), and Potato virus Y (PVY).

Jiang et al. ^[20] developed a rapid detection colloidal gold test strip for Tomato zonate spot virus (TZSV), which can obtain the detection result within 5 minutes. In 2021, the patent for the kit for detecting crop viruses and the method and application for detecting crop viruses applied for by Sun et al. ^[21] was publicly announced online on 2021. This invention can be applied to the detection of viruses in herbaceous and woody crops. On September 2, 2023, a patent filed by Wang et al. ^[22] for a PCR kit for the simultaneously detecting Tomato chlorosis virus (ToCV), Southern tomato virus (STV), and Tomato yellow leaf curl virus (TYLCV) was disclosed. On May 3, 2024, the patent for a real-time fluorescence quantitative PCR kit for detecting Tomato brown rugose fruit virus (ToBRFV) applied for by Li et al. ^[23] was announced. On September 6, 2024, the patent for an RT-PCR kit for detecting brown stripe pathogen in sugarcane applied for by Shan et al. ^[24] waspublished. The emergence of these recent studies reflects the great interest and widespread attention of the Chinese scientific community in crop disease kits.

3.Classification and Applications of Crop Disease Rapid Test Kits

3.1 Classification

According to different analytical methods, crop disease kits are mainly divided into five categories: ELSIA kits, RT-PCR kits, PCR kits, lateral flow immunoassay kits, and colloidal gold chromatography test strips. Among them, some ELSIA kits, lateral flow immunoassay kits, and colloidal gold chromatography test strips have simple operation methods and do not require trained professionals to operate. These are the main categories of crop disease kits currently sold in the terminal agricultural input retail markets abroad.

3.2 Applications

Currently, the crop disease rapid test kits from Agdia Inc. (US), Pocket Diagnostic (UK), and Vetall Laboratories International Veterinary Company (South Korea) have been launched into the terminal market, and their main products and properties are listed in Table 1. These test kits can provide detection results within 5 to 10 minutes, and farmers can quickly learn how to use them through the operation instructions. At present, the main test kits on the market mainly include Erwinia amylovora test kit, Fusarium oxysporum test kit, Ralstonia solanacearum test kit, Phytophthora test kit, Alfalfa mosaic virus (AMV) immunostrips, Tobacco streak virus (TSV) immunostrips, Zucchini yellow mosaic virus (ZYMV) immunostrips, and so on. These crop disease rapid test kits are convenient to carry, simple to operate, highly sensitive, and have low requirements for storage conditions. Users can quickly learn how to use them by reading the product instructions, without the need to purchase expensive equipment. They can be stored at 2-30°C for two years, and within 10 minutes, it can be determined whether the crop is infected with the pathogen, so as to determine whether pesticides need to be sprayed and which pesticides should be sprayed. Since different causes may lead to similar disease characteristics in crop diseases, to meet the market demand, South Korean kit manufacturers have provided detailed descriptions of disease symptoms according to the applicable situations of different kits, so that users can understand which kit should be selected for testing. Figure 1 shows the composition of a test kit manufactured in South Korea. If a crop shows disease symptoms, growers or agronomists can pre-judge the type of kit to be selected according to the appearance characteristics of the crop. For example, if symptoms of Fusarium oxysporum infection appear, a rapid kit for Fusarium oxysporum can be selected for detection. The changes before and after the test kit assay are presented in Figure 2. If the test result is positive, the following pesticides can be selectively used for spraying and treating the diseased crop: Thiophanate-methyl 70% WP, and Pyraclostrobin 250 g/L EC, or Carbendazim 80% WDG and Difenoconazole 40% SC. If a positive result is detected by the rapid kit for Ralstonia solanacearum, one of the following pesticides can be selectively used for spraying and treating the diseased crop: Bacillus subtilis 10 billion CFU/g WP, 300 million CFU/g Trichoderma harzianum WP, Zhongshengmycin 3% WP, or Metalaxyl 5%+ Thiram 33% WP. The official website information of Pocket Diagnostic in the United Kingdom shows that its customer, Potadaho Seed Services in the United States, has used the Potato virus Y (PVY) kit and the Phytophthora Test kit in Nevada, California, Idaho, eastern Oregon, and Montana. The early use of these kits has well helped farmers avoid yield losses. Luis Gustavo Orellana Aragon, a customer of Pocket Diagnostic from Guatemala, used the bacterial wilt kit in greenhouses at the request of his local customers. He reported that the early use of the kit can help geranium farmers ensure the yield and achieve a bumper harvest.

No.	Trade name (Trade mark)	Manufacturer	Application range	Pathogen types
1	ImmunoStrip®	Agdia, Inc. (USA)	corn and other gramineous crops	Maize dwarf mosaic virus (MDMV)
2	Pocket Diagnostic®	Pocket Diagnostic (UK)	Potato	potato virus YO (common strain) and potato virus YN (necrotic strain)
3	SensPert TM	Vetall Laboratories Internation- al Veterinary Company (Korea)	Tomato eggnlant hot nenner	Fusarium oxysporum /Ralsto- nia solanacearum

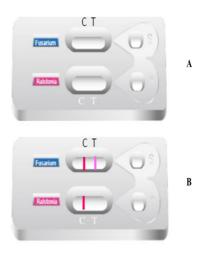
Table1: Main manufacturers and products of crop disease rapid Test kits

Many institutions and university laboratories in China have achieved a lot of research results on crop disease kits, such as the beginning of this paper mentioned Zhou Xueping team developed dot-ELISA crop virus rapid detection kit is mainly used in agricultural regulatory agencies and customs inspection and quarantine. However, most of these kits are limited to inspection and quarantine and scientific research, and there is no successful case of cooperation between enterprises and research institutes to promote these kits to the end market. At present, some companies have imported some crop disease detection products as agents. However, the products are mainly used in customs inspection and quarantine, scientific research institutions and universities for experimental research. Simple, inexpensive and portable crop disease rapid kits for on-site testing in farmers' fields have not yet been pushed into the terminal retail market.

Figure 1: Composition of a kit produced by Vetall Laboratories International Veterinary Company (South Korea) A: instructions; B: test device; C: extraction buffer; D: disposable spoon; E: disposable dropper



Figure 2: The changes before and after the test kit assay A: before assay; B: after assay



4. Problems and Countermeasures of Crop Disease Rapid test Kits in China

Although crop disease rapid test kits for farmers have many advantages, there are significant difficulties in promoting them domestically. The author analyzes that the main reasons are as follows: Firstly, pesticide enterprises are the most suitable for promoting such products. However, the application of these products will reduce the use of pesticides, which will inevitably affect the pesticide sales volume of pesticide manufacturers. Sales personnel are reluctant to promote these products due to concerns about their own performance and income. Secondly, there is a wide variety of bacterial and viral pathogens in crops. Enterprises need to invest time and capital to develop kits targeting different bacteria, fungi, and viruses. The early research, development, promotion, and publicity stages require substantial investment in human resources, financial resources, and material resources. Few enterprises are willing to invest the time and effort in researching, developing, and promoting such products. Finally, people have a low acceptance of new things. Most individuals and enterprises are reluctant to be the "first to take the plunge." The difficulties in researching, developing, and promoting crop disease rapid test kits in the terminal agricultural input market have deterred many enterprises.

The crop disease rapid test kits are products that can detect early signs of crop diseases and guide users to use pesticides appropriately according to the types of detected diseases. They can help farmers nip crop diseases in the bud. The rational use of crop disease rapid test kits can not only reduce the amount of pesticides used, decrease the waste and pollution of pesticides, but also indirectly reduce pesticide residues in agricultural products and make food safer. At present, facing problems such as low marketization and difficulty in promotion of these products, government departments are needed to guide, encourage, and support enterprises to cooperate with university laboratories or scientific research institutions in jointly researching, developing, and promoting such products. The research and application of these products are in line with the Action Plan for Zero Growth of Pesticide Use by 2020 issued by the Ministry of Agriculture of China ^[25], and also conform to the strategies and concepts of agricultural sustainable development, green development, and rural revitalization. The government can selectively identify and cultivate a few potential and strategic enterprises, incorporate the research and development projects of crop disease rapid test kits into the rural revitalization strategy, and let a small number of enterprises take the lead in developing such projects. Once these enterprises taste the benefits brought by the projects, it is believed that more enterprises will scramble to imitate and join the army of promoting crop disease rapid test kits.

5. Prospects Analysis of Crop Disease Rapid test Kits in China

China has always paid great attention to food safety. There are many technologies and methods for detecting pesticide residues in vegetables and fruits. Some enterprises have been fined due to excessive pesticide residues, and some food enterprises have been prohibited from exporting their goods due to the same problem. The crop disease rapid test kits can help growers detect crop diseases at an early stage and control and reduce the use of pesticides, ensuring the safety of plant-derived food from the source. It is believed that after these products become popular in the terminal agricultural input retail market, food retail stores will rarely be troubled by excessive pesticide residues in their products, and food exporters will significantly reduce the situations where they cannot fulfill their contractual obligations due to excessive pesticide residues.

Currently, the whole world is pursuing a low-carbon, green, and environmentally friendly lifestyle. The research, development, promotion, and application of crop disease rapid test kits can enable farmers to reasonably reduce the use of pesticides while ensuring a bumper harvest, which will also indirectly reduce pesticide production. At present, there are not many enterprises globally engaged in researching and selling the crop disease rapid test kits. This is a good opportunity for developing and commercially promoting such kits at the terminal level. It is hoped that more technology-based enterprises will cooperate with university laboratories or scientific research institutions to engage in the industrial production of such products as soon as possible to seize the market opportunity. As the 'crop doctor's eyes' of the crop disease rapid test kit, it is believed that in the near future, with the guidance and encouragement of the government, enterprises will perceive the market potential and value of crop disease rapid detection kits and enter this industry. With the development and standard improvement of on-site crop disease rapid test kits in the fields, the subsequent market will become standardized. At that time, the ultimate beneficiaries will not only be enterprises, farmers, and growers, but also the vast number of consumers, because

less pesticide spraying will make the food flowing to the terminal market safer and healthier.

Funding

no

Conflict of Interests

The author(s)declare(s) that there is no conflict of interest regarding the publication of this paper.

References

- Oerke, E. C.: Crop losses to pests. Journal of Agricultural Science, 2006, 144(1):31-43. DOI: 10.1017/ S0021859605005708.
- [2] Savary, S., Willocquet, L., Pethybridge, S. J.: The global burden of pathogens and pests on major food crops [J/OL]. Nature Ecology & Evolution, 2019(3): 430-439. DOI: https://doi.org/10.1038/s41559-018-0793-y.
- Zhou, H., Tang, Z., Zhao, L.: Rapid Detection of Four Major Crop Viruses. The Beijing News, 2021-11-08. https://www. bjnews.com.cn/detail/163635508014893.html.
- [4] Ministry of Agriculture and Rural Affairs of the People's Republic of China. Decree of the State Council of the People's Republic of China No. 725: Regulations on the Prevention and Control of Crop Diseases and Pests. (2020-4-3)[2024-9-11] http://www.zfs.moa.gov.cn/flfg/202004/t20200403_6340771.htm.
- [5] Dietzgen, R. G., Sander, E.: Monoclonal antibodies against a plant virus. Arch Virol. 1982(74):197-204.
- [6] Diaco, R., Hill, J. H., Hill, E. K.: Monoclonal antibody-based biotin-avidin ELISA for the detection of Soybean mosaic virus in soybean seeds. Journal of General Virology. 1985, 66(10): 2089-2094. DOI: https://doi.org/10.1099/0022-1317-66-10-2089.
- [7] Richard, K. L., Sally A. M., David, G.: Kit for diagnosing plant diseases: 5047207. 1991-9-10.
- [8] Richard, K., Sally A. M., David, G.: Method and a kit for diagnosing plant diseases: 5219761. 1993-6-15.
- [9] Kim, J. H., Yang, M. Y., Kim, J. M.: Simultaneous diagnostic method for the ralstonia solanacearum and fusarium oxysporum using semi-quantitative lateral flow immunoassay: 10-2154628. 2020-09-04.
- [10] Razo, S. C., Safenkova, I. V., Drenova, N. V.: New lateral flow immunoassay for on-site detection of Erwinia amylovora and its application on various organs of infected plants. Physiological and Molecular Plant Pathology, 2021, 114: Article101637.
- [11] Wei, M., Xiang, N., Zhang, Z.: Detection of Grapevine Fanleaf Virus and Potato Virus Y by Biotin labeled cDNA Probe of Coat Protein Gene. Acta Phytopathologica Sinica., 1995, 25(4):331-334.
- [12] Zhang, J., Liu, Z., He, P.: Diagnosis of Citrus Huanglongbing by Monoclonal Antibody-Colloidal Gold Method and PCR Technology. Scientia Agricultura Sinica, 1997, 30(5): 94-96.
- [13] Li, M., Wei, M., Zhu, S.: Brief Report on the Development of Alkaline Phosphatase Enzyme-Linked Diagnostic Kits for Tobacco Ringspot Virus and Potato Virus X. Plant Quarantine, 2001, 2(15): 89.
- [14] Wang, L., Wu, X., Xiang, N.: Gene Chip, Nucleotide Sequence and Kit for Detecting Viruses in Solanaceous Crops: 200610081221.1. 2007-04-25.
- [15] Wang, L., Wu, X., Xiang, N.: Gene Chip, Nucleotide Sequence and Kit for Detecting Viruses in Leguminous Crops: 200610081219.4. 2007-04-25.
- [16] Wei J. T., Liu, H. X., Liu, F.: Miniaturized paper-based gene sensor for rapid and sensitive identification of contagious plant virus. ACS Applied Materials & Interfaces, 2014, 6(24): 22577-22584. DOI:dx.doi.org/10.1021/am506695g.
- [17] Tian, J.: Preparation of Monoclonal Antibodies against Tomato Zonate Spot Virus and Development of Colloidal Gold Immunochromatographic Test Strips. Nanjing: Nanjing Agricultural University, 2015.
- [18] Li, G., Ma, J., Kong, J.: Development of Antibodies against Potato Virus V and TAS-ELISA Kit. Journal of Inspection and Quarantine, 2016, 26 (5): 5-8.
- [19] Ruan, X., Deng, H., Wang, X.: Development and Application of Colloidal Gold Test Strips for the Detection of Multiple Tobacco Viruses. Tobacco Science & Technology, 2018, 51(10): 33-38.

- [20] Jiang, N., Xia, Z., Lu, C.: Development and Application of a Rapid Detection Strip for Tomato Zonate Spot Virus in Tobacco. Tobacco Science & Technology, 2023, 56(11): 10-15,53.
- [21] Sun, L., Wang, L., Shang, Q.: Kit for Detecting Crop Viruses, Method for Detecting Crop Viruses and Applications Thereof: 202111076207.3. 2021-11-9.
- [22] Wang, H., Gao, M., Li, J.: Multiplex PCR Primer Compositions, Detection Methods, and Detection Kits for Simultaneously Detecting Three Tomato Viruses: 202310820008.1. 2023-09-22.
- [23] Li, T., Wang, J., Yang, J.: Real time Fluorescent Quantitative PCR Primer, Kit and Application for Detecting Tomato Brown Rugose Fruit Virus: 202410314214.X. 2024-05-03.
- [24] Shan, H., Huang, Y., Gao, Si.: A Kit, Detection Method and Application for Detecting Brown Stripe Pathogen in Sugarcane: 202410885114.2. 2024-09-06.
- [25] Ministry of Agriculture and Rural Affairs of the People's Republic of China. Decree No. 725 of the State Council of the People's Republic of China: Notice of the Ministry of Agriculture on Issuing the Action Plan for Achieving Zero Growth in the Use of Chemical Fertilizers by 2020 and the Action Plan for Achieving Zero Growth in the Use of Pesticides by 2020.(2017-11-29). http://www.moa.gov.cn/nybgb/2015/san/201711/t20171129_5923401.htm