Recent advances in E-monitoring of Plant Diseases

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Abstract

Infectious plant diseases are caused by pathogenic microorganisms, such as fungi, oomycetes, bacteria, viruses, phytoplasma, and nematodes. Plant diseases have a significant effect on the plant quality and yield and they can destroy the entire plant if they are not controlled in time. To minimize disease-related losses, it is essential to identify and control pathogens in the early stages. Plant disease control is thus a fundamental challenge both for global food security and sustainable agriculture. Conventional methods for plant diseases control have given place to electronic control (E-monitoring) due to their lack of portability, being time consuming, need for a specialized user, etc. E-monitoring using electronic nose (e-nose), biosensors, wearable sensors, and 'electronic eyes' has attracted increasing attention in recent years. Detection, identification, and quantification of pathogens based on electronic sensors (E-sensors) are both convenient and practical and may be used in combination with conventional methods. This paper discusses recent advances made in E-sensors as component parts in combination with wearable sensors, in electronic sensing systems to control and detect viruses, bacteria,

pathogens and fungi. In addition, future challenges using sensors to manage plant diseases are investigated.
Keywords: E-sensing, Electronic head, Biosensor, Disease control, Wearable sensor.

1. Introduction

During the last century, the world population increased exponentially from 1.6 billion people in 1900 to 7.7 billion in 2019. With the current rates of population growth, it is predicted that in 2050 the world population will approach 10 billion, which will lead to a huge rise in the consumption of resources. In this context, it is calculated that to feed the world population, crop production will have to increase by 60-100% by 2050, and this simultaneously with the reduction of environmental damage caused by agriculture (Emadi & Rahmanian, 2020; Hunter et al., 2017; Struik & Kuyper, 2017). Therefore, the current rate of population growth makes it essential to develop new strategies for sustainable food production (Poveda, 2021).

To increase crop production, the development of new technologies in the agricultural sector is essential. This includes the development of new forms of agriculture, such as vertical farming or controlled-environment culture (Benke & Tomkins, 2017), together with the implementation of precision agriculture (Vuran et al., 2018) controlled by artificial intelligence (AI) (Jha et al., 2019) (Pathan et al., 2020). Therefore, the agriculture of the future will have to face important challenges based on increasing crop productivity while maintaining environmental sustainability (Tian et al., 2021).

One of the main threats to increasing crop productivity is the losses caused by the attack of pathogens. Phytopathology is the study of plant diseases through their detection, the understanding of host-pathogen interactions, and the development of strategies to reduce the agricultural losses they cause (Mitra, 2021). Together, pathogens, animals and weeds annually cause losses in world crops between 20 and 40% of global agricultural productivity, of which between 10 and 15% are direct losses caused by pathogens (Savary et al., 2012) (Mitra, 2021). Specifically, plant pathogens are responsible for annual economic losses of around $220 billion for the agricultural sector, causing problems of access to food for more than 800 million people (Mitra, 2021).

Plants affected by pathogens suffer from various plant diseases that cause poor yields in terms of both quantity and quality, by affecting their roots, stem, shoots, leaves, flowers, and fruits. The main microorganisms that cause disease in crops are viruses, bacteria, fungi, oomycetes and nematodes as shown in Table 1, indicating the species of greatest agricultural importance.

Table 1. Main agricultural pathogens

Group	Species	Reference	Group	Species	Reference	Group	Species	Reference
Viruses	*Tobacco mosaic virus* *Tomato yellow leaf curl virus* *Tomato spotted wilt virus* *Cucumber mosaic virus* *Cauliflower mosaic virus* *Potato virus Y* *African cassava mosaic virus* *Plum pox virus* *Potato virus X* *Bromemosaic virus* *Citrus tristeza virus* *Potato leafroll virus* *Barley yellow dwarf virus*	Scholthof et al., 2011	Bacteria	*Pseudomonas syringae* *P. savastanoi* *Xanthomonas oryzae* pv. *oryzae* *X. campestris* *X. axonopodis* *Ralstonia solanacearum* *Erwinia amylovora* *Dickeya dadantii* *D. solani* *Xylella fastidiosa* *Pectobacterium carotovorum* *Candidatus* Liberibacter asiaticus *Clavibacter michiganensis* *Magnaporthe oryza* *Puccinia* spp.	Mansfield et al., 2012	Nematodes	*Heterodera* spp. *Globodera* spp. *Meloidogyne* spp. *Pratylenchus* spp. *Ditylenchus dipsaci* *Radopholus similis* *Bursaphelenchus xylophilus* *Rotylenchulus reniformis* *Nacobbus aberrans* *Xiphinema index* *Aphelenchoides besse*	Jones et al., 2013
Oomycetes	*Phytophthora infestans* *P. ramorum* *P. sojae* *P. capsica* *P. parasítica* *P. cinnamomi* *Plasmopara vitícola* *Pythium ultimum* *Albugo candida*	Kamoun et al., 2015	Fungi	*Botrytis cinerea* *Fusarium graminearum* *F. oxysporum* *Mycosphaerella graminicola* *Blumeria graminis* *Colletotrichum* spp. *Melampsora lini* *Ustilago maydis* *Verticillium dahlia* *Phakopsora pachyrhizi*, *Rhizoctonia solani*	Dean et al., 201			

Detecting the presence of the pathogen in the crop as soon as possible minimizes the losses that may occur in productivity, allowing a targeted and specific treatment. In this sense, there are different methods of detecting diseases in crops, grouped into direct methods, if they detect the presence of the pathogen, or indirect methods, if they detect the effects they cause in plants. Within the direct methods, molecular and biochemical tools are currently used, such as polymerase chain reaction (PCR), fluorescence *in situ* hybridization (FISH) for bacterial detection, enzyme-linked immunosorbent assay (ELISA), immunofluorescence (IF), or flow cytometry (FCM). However, pathogen detection can sometimes deliver erroneous data, as it does not differentiate living microorganisms from components. For example, the detection of viruses, the use of methodologies that detect in the same way virion particles and their scattered components (proteins or nucleic acids). Furthermore, the protocol to be carried out for the detection of each of the different plant pathogens can be very different and complicated, also vary according to the affected crop. This may significantly complicate the testing procedure. On the other hand, indirect methods analyze parameters such as morphological change, transpiration rate change, temperature change, and the volatile organic compounds released by infected plants. Such methods include thermography, hyperspectral techniques, fluorescence imaging, or gas chromatography (Fang & Ramasamy, 2015) (Martinelli et al., 2015). In addition, biosensors, e-nose, optical-based sensors and wearable sensors provide indirect assays for e-monitoring of plant disease management that are discussed in the following sections. Therefore, the use of new electronic and information technologies in crop production will allow agriculture to meet the future challenges of rapidly predicting plant diseases and acting effectively in a targeted manner, reducing the use of chemical pesticides and improving environmental sustainability.

2. Importance of E-monitoring

Reducing plant losses due to environmental stresses and pathogens, improving resources utilization efficiency, and selecting optimal plant traits are major challenges in the field of agricultural industries across the world. The ability of e-monitoring to provide accurate quantitative and qualitative identification of plant pathogens would be a turning point in plant pathology. Reliable and accurate identification of organisms responsible for a plant disease is the main prerequisite for implementation of disease management strategies (Ray et al., 2017).

Since many plant pathogens cause similar symptoms, it is important to be able to distinguish between different species using e-monitoring. Conventional methods for plant disease management are not usually sensitive enough and are off-site, expensive, time-consuming, and need trained personnel. The need for a sensitive, specific, user-friendly and rapid methods, providing on-site sampling for plant disease management is becoming increasingly apparent (Fang & Ramasamy, 2015). E-monitoring with the aid of intelligent plant sensors connected to electronic devices improves plant productivity (Lee et al., 2018). Therefore, new technologies are needed to accurately monitor the physiological and growth responses of plants to the environment in real time with high spatial and temporal resolution (Giraldo et al., 2019).

Wearable technology composed of biosensors, gas sensors, and optical sensors embedded in clothes or worn as accessories on the body have been extensively studied in biomedical applications to continually track the health or fitness-related biometric information. Such wearable technology has also been considered as an emerging tool for the electronic control and detection of plant diseases. Due to the ability of these tools to continuously track important physiological and pathological parameters *in situ*, as well as remote control and communication in plants, they have attracted attention in recent years.

Plant wearables have the ability to achieve simple, accurate and continuous monitoring of plant health at large-scale compared to IR fluorescence-based nanobionics and/or Raman spectroscopy, which often require sophisticated tools and require off-site analysis (Yin et al., 2021). Thin-film plant wearable (TFPWs) have the ability to bind to irregular plant tissue surfaces due to their noninvasive and flexible properties. TFPW is suitable to monitor environmental parameters such as temperature, moisture, and biological parameters such as water potential, plant tissue displacement/strain and volatile organic matter composition. For example, (Lee et al., 2014) developed an all-carbon film based on field-effect transistor (FET) sensors using single walled carbon nanotubes (SWCNTs) that can wirelessly measure the concentrations of toxic gases of plants down to part-per-million (ppm) sensitivity. The results of their study showed that these flexible sensors have excellent mechanical properties and good adhesion to nonplanar surfaces of biological materials, which show a unique potential for both wearable electronic sensors and bio-implantable sensors.

3. Electronic Sensing Systems

As a new strategy, an electronic agricultural sensing system (EASS) using interconnected individual sensors (Fig 1) to form a composite intelligent biosensing system which is analogous

to an artificial nervous system and brain. So far, this concept has not been used in agriculture. Such an electronic sensing system uses individual artificial sensors including electronic tongue (biosensors), e-nose, electronic eye, and electronic ear (acoustic-based sensors) connected together and has been termed 'the artificial head' (Wide, 2001). It may be possible to add other wearable sensors to such a composite sensor system e.g. e-touch (haptic sensing) as electronic 'skin' and adapting the concept for agricultural use.

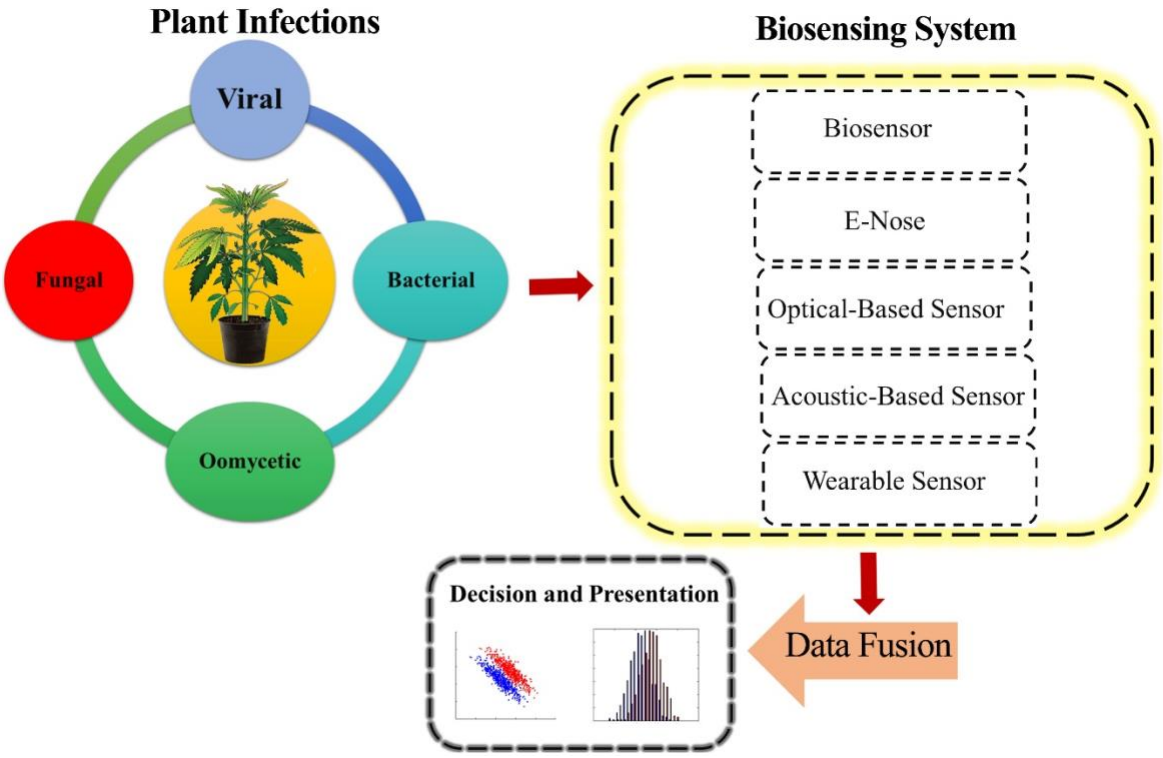

Fig 1. Application of biosensing systems for plant infection sensing

The chewing process can also be considered in this regard for which a significant application has not yet been defined in agriculture. In general terms an EASS could include the chewing process, as a video recording system to identify a given sample, employing an artificial sensor 'mouth' to measure the crushing and chewing process of a sample, an e-nose to measure the emitted odour, an electronic ear to determine the resistance to chewing and to hear the sound of crushing, and an e-tongue to measure the food taste (Wide et al., 1999).

By combining data from the individual sensors into a system, we can obtain more useful information compared when we obtain the response from each sensor separately. Data fusion uses different information sources to better understand the response of individual sensors (Sundic et al., 2000). In this paper, for the first time, the concept of EASS is proposed for use

in identifying plant diseases based on the following biosensors: e-nose, electronic eye, acoustic-based sensors, combined with wearable sensors. These concepts are discussed in the following sections.

4. Different types of biosensors for plant disease sensing

4.1. E-nose

The electronic nose (e-nose), which is also called artificial nose or artificial olfaction system, is an apparatus that simulates human olfactory senses (Sun et al., 2019; Zhong, 2019). An e-nose system is a sensor based intelligent tool that is designed to detect and discriminate complex odors using an array of non-selective sensors (Jiarpinijnun et al., 2020; Santos et al., 2019). A typical e-nose system consists of three main elements including: sample handling and odour delivery system, detection system (sensor array), and data processing and machine-learning algorithms (Long et al., 2019; Rahman et al., 2020)

The sensor array is composed of different gas sensors treated with different odour-sensitive biological or chemical materials to be sensitive to different substances (Boeker, 2014; Kiani et al., 2016). Different types of gas-sensitive sensors including metal oxide semiconductor (MOS) sensors, quartz crystal microbalance (QCM) sensors, surface acoustic wave (SAW) sensors, electrochemical (EC) sensors, optical sensors, calorimetric sensors, and biomimetic sensors can be employed in e-nose systems (Bonah et al., 2020; Gliszczyńska-Świgło & Chmielewski, 2017; Sanaeifar et al., 2017).

When a gas, from the headspace of a sample, comes in contact with individual sensors in the sensor array, the sensor produces a signal that is proportional to the concentration of the substance detected (Deshmukh et al., 2015; Jasinski et al., 2017). This signal is typically a change in the output voltage of the sensor. The sensor reaction signals are recorded and converted into digital data by the signal processing system to generate a database that is subjected to multivariate analysis, to identify and characterize the experimented odour (Peris & Escuder-Gilabert, 2016). The voltage variation signals are loaded in processing system. Different numerical characteristic parameters can be extracted from these curves and analyzed. These features include : maximum sensor response, sensor impregnation time, sensor recovery time, mean and maximum slopes of the sensor response, and area under curve (Huang et al., 2017; Ordoñez-Araque, 2020). Differently supervised and unsupervised chemometrics can be applied on the multidimensional output data of sensor arrays to provide precise and predictive results (Li et al., 2020). These statistical analysis approaches include Principal Components

Analysis (PCA), Artificial Neural Networks (ANNs), Linear Discriminant Analysis (LDA), Partial Least Square Regression (PLSR), and Support Vector Machines (SVM), which are fully described in a reviews by (Di Rosa et al., 2017; Karakaya et al., 2020).

The organic volatile compounds (VOCs) released by agricultural material can be detected by e-nose system and used for various applications in agricultural and food related fields. Accordingly, several literature reviews have revealed the applications of e-nose in the agro-food arena (Ali et al., 2020; Jia, Liang, Jiang, et al., 2019; Roy & Yadav, 2021; Shi et al., 2018). In the case of plants, which are among the organisms emitting the highest diversity of VOCs (Tholl et al., 2021), the released VOCs serve excellent information about plants health status (Effah, 2020), since the composition of emitted VOCs by plants is changed regarding of the type of biotic or abiotic stresses that the plant is exposed to (Cellini et al., 2018). The specific and characteristic VOCs that are emitted by pathogenic microorganisms provide unique odour fingerprints that can be used as biomarkers for pathogen identification and discrimination (Bonah et al., 2020; Moisan et al., 2019). Although the e-nose system cannot quantify the various volatile compounds in a plant headspace gas, the sensor array of e-nose setup can generate a unique profile regarding the composition of the VOCs generated by a specific pathogen or pest infected plant (Fig 2). The system can be then trained to discriminate between healthy and infected plants or even among multiple infections induced by different agents in a non-destructive manner based on only the sample odour (Laref et al., 2019).

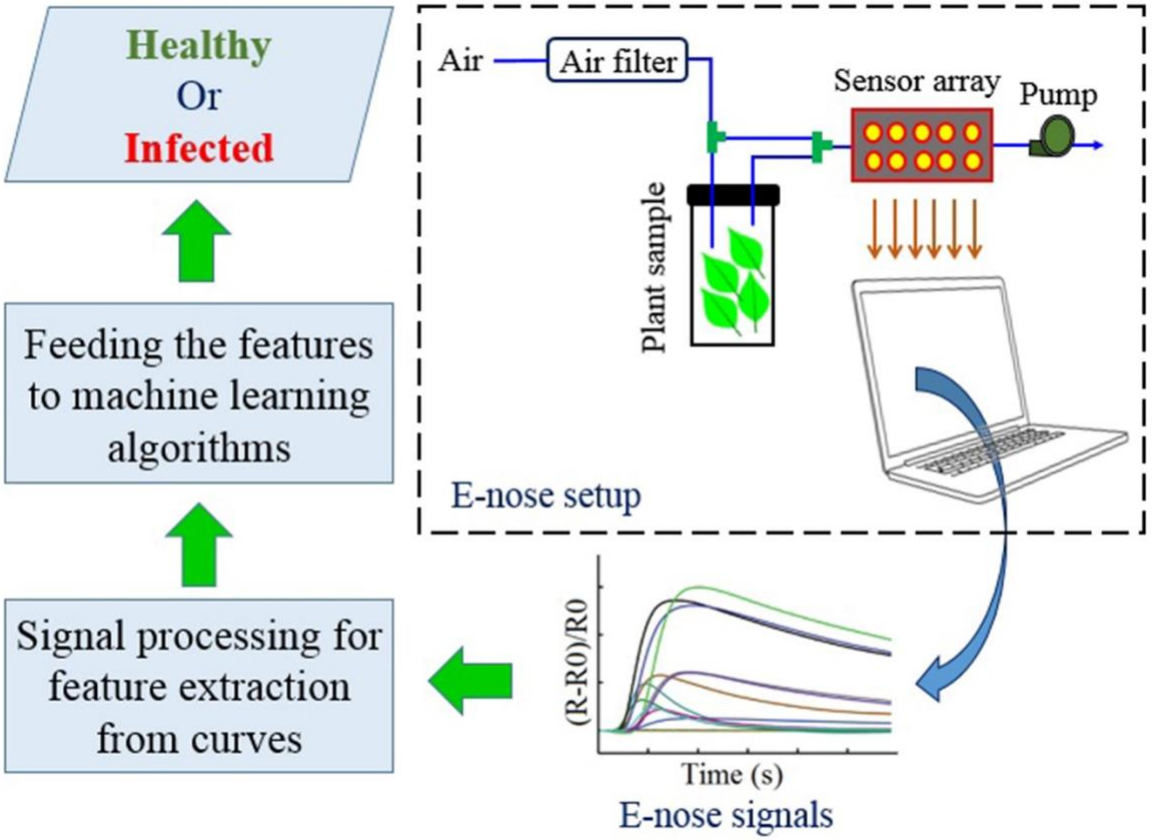

Fig 2. A typical e-nose system for plant disease monitoring

An e-nose system having ten different MOS sensors was used by Chang et al. (2014) for detection of plant pathogenic bacteria on chilli and papaya plants. The chilli samples were infected by three different bacteria including *E. carotovora*, *R. solanacearum*, and *X. campestris pv. vesicatoria*, meanwhile the papaya samples were infected by two types of bacteria namely *Rickettesia sp* and *Erwinia s*p. PCA showed that the sensor data was significant toward discrimination of the odours emitted by plants infected by different bacteria.

An electronic nose system was applied by Jiarpinijnun et al. (2020) for early monitoring of *Aspergillus* fungus contamination on Jasmine brown rice grains. PCA, LDA and SVM classifiers were used to discriminate different levels of contamination and PLS model was used for fungal growth prediction. It was reported that an e-nose is an accurate technique for early detection of fungal infection on rice grains prior to visual symptoms emergence.

In another study, the application of e-nose system was evaluated for detecting the *Citrus Tristeza virus* infection in mandarin orange. A classification accuracy of 97.58% was obtained for differentiating between thee healthy and infected samples using adaptive boost ensemble of decision tree classifier (Hazarika et al., 2020).

A study was conducted by Borowik et al. (2021), in which an e-nose was successfully employed for distinguishing between two important forest pathogenic oomycetes namely *Pythium* and *Phytophthora* using SVM classification model.

A summary of some studies on plant disease detection using e-noses is presented in Table 2. It should be stated that most of research studies in this arena are about detection of bacterial or fungal infections, while diseases with other agents have been less focused. There are also good review articles that have provided detailed information about the e-nose and its applications in plant disease detection (Bonah et al., 2020; Cellini et al., 2017; Wilson, 2018), showing great potential of e-nose systems to be developed and used for non-destructive and early stage detection of plant diseases.

Table 2. Summary of e-nose applications for plant disease detection.

Disease name	**Disease agent**	**E-nose system**	**Most effective sensors**	**VOC**	**Acquisition time (s)**	**Modeling algorithm**	**Reference**
Potato brown and ring rot	Bacterial	PEN3	-	Alcohols and aliphatic substances	60	PCA, LDA	(Biondi et al., 2014)
Fire blight and blossom blight in apple plant	Bacterial	EOS507C, PEN3	-	hexenal isomers, 2,3-butanediol, 3-hydroxy-2-butanone, phenylethyl alcohol	EOS507C: 180, PEN3: 120	PCA, LDA	(Cellini et al., 2016)
Soft-rot infection in potatoes	Bacterial	WOLF 4.1	-	carbon monoxide, ethylene oxide, nitric oxide	120	PCA , C5 decision tree	(Rutolo et al., 2018)
Lethal Bronzing Disease in Cabbage	Bacterial	Laboratory developed setup	MQ5 , MQ8	Hydrogen	-	PCA	(Oates et al., 2020)
Detection of potato tuber	Bacterial	Laboratory	TGS2620	Alcohol, organic solvents	120	RBFNN, SVM	(Chang et al., 2017)

soft rot disease *Botrytis* sp., *Penicillium* sp. and *Rhizopus* sp. in ripe strawberry fruits	Fungal	developed setup PEN3	W1C, W3C, W1S, W2S , W3S	Ammonia, aromatic compounds, methane, alcohols	60	PCA, ANN	(Pan et al., 2014)
B. cinerea infestation of tomato plant	Fungal	PEN3	W1A, W5B	2-Carene, β-Phellandrene , 2-ethyl-1-Hexanol, Nonanal, Naphthalene	70	PCA, LDA, MLR	(Sun et al., 2018)
Aspergillus spp. contaminatio n in rice	Fungal	PEN2	W5B, W2B, W1B, W2C	Nonanal, nonadecane, cis-Thujopsene, 1-octanol, 1-octen-3-ol and diethyl phthalate	90	PCA, BPNN, SVM, LVQ, PLS	(Gu et al., 2019)
Penicillium expansum and *Aspergillus niger* in apples	Fungal	PEN2	W5S, W1S, W1W, W2S, and W2W	Oxynitride, Aromatic compounds, Sulfides , organic sulfides, methane, alcohols	150	LDA, SVM, BPNN, RBFNN	(Jia, Liang, Tian, et al., 2019)

4.2. Electrochemical Biosensors

Common methods for detection and identification of fungal pathogens that cause disease in plants rely mainly on morphological, microbiological and biochemical identifications. Traditional methods are not sensitive enough, and therefore new methods have been developed in the last decade for identifying plant pathogens (Zhao et al., 2020). Over recent years,

biosensors have received special attention due to their promising results in identification, classification, detection, and quantification of plant diseases. Therefore, biosensors provide rapid and accurate plant disease diagnosis, reduce the prevalence of diseases, and effectively facilitate disease management (Ali et al., 2021). A sensor is a device that converts chemical information, such as the concentration of a particular sample into an analytical signal. The electrochemical method uses particular aspects of an electrochemical biosensor. In addition to the electrochemical method, the sensor signal readout format provides other aspects of biosensor-based methods for pathogen detection. Electrodes can be fabricated from several materials through fabrication processes (Berto et al., 2019).

A "Biosensor" is therefore a device designed to measure the presence and amount of a particular biological substance through combining a mechanism that converts the physicochemical changes appearing in response to the analytes present in the sample with a receptor which is capable of manifesting this as a recognizable signal (Hong & Lee, 2018). Due to their ability in selective and rapid detection of only analyzed materials, biosensors are actively used in many industrial fields such as medical diagnosis, new drug development, plant pathology, food safety testing and environment monitoring. Moreover, the development of key supportive elements such as various interfaces and nanotechnology has increased the application range of biosensors to point-of-care tests and Internet-of-things (IoT) (Alonso et al., 2020). The transduction element of electrochemical biosensors is an electrochemical cell whose main component is a working electrode. A three-electrode system (including working, auxiliary, and reference electrodes) is usually used in potentiostatic system, while the two-electrode format (working and auxiliary) is often used for conductometry and electrochemical impedance spectroscopy. An electrode is an electronic conductor in which the charge is transported by the movement of electrons and/or holes. Therefore, electrodes may be fabricated from conductors and semiconductors, including metals, like e.g., gold, and nonmetals, like e.g., carbon. The materials, fabrication methods and designs used for electrode fabrication affect the structure and properties of electrode, which result in the performance characteristics of the biosensor. The most important functional properties of biosensors are their sensitivity, selectivity, limit of detection (LOD) and dynamic range. Requirements for these affect the biosensor's fabrication cost, manufacturability, disposability and measurement capability (Kumar & Arora, 2020).

Over recent years, several studies have been reported on the development of biosensors for detection of plant diseases. Among the most important citrus viruses we can refer to *citrus tristeza virus* (CTV), which has caused high plant mortality, especially in grafted citrus seedlings worldwide (Harper & Cowell, 2016). Plants infected with CTV may show mild to severe symptoms depending on the type of virus and the sensitivity of different types of plants. Tables 3-5 show the summary results for the use of biosensors to identify plant fungi, oomycetes, bacteria, and viruses.

Umasankar and Ramasamy (2013) reported a nanomaterial-based electrochemical sensor for plant disease diagnosis. Gold nanoparticles (GNP)-modified cathode was used for electrochemical detection of methyl salicylate, a major volatile organic compound released by plants during infection (Umasankar & Ramasamy, 2013). In DNA based electrochemical strategies, voltametric methods have been used for discriminative investigation of nucleic acid structure and its modification with the simultaneous identification of all DNA bases without any need for hydrolysis phase (Patel, 2021). Adding newer sensors for diagnosis of disease e.g., fibre-optic biosensors (FOBS) and electrochemical biological sensors is a more useful way to overcome uncertain diagnosis of plant infections. Considering the importance of monitoring plant health and the current status of nano-biosensors, and given the general drawback in existing techniques, advances made in nanomaterials and new biomarkers, will provide an appropriate impetus to researchers to switch their focus to growth monitoring and plant health.

Table 3. Different biosensors proposed for fungi and oomycete pathogen detection

Type of pathogen	Sensing platform	Organism	Nano material	Signal mode	LOD	Range	Analysis time	Reference
Oomycete	SPCE	*Phytophthora cactorum*	SnO_2	CV	82 nM	0.6 µM–0.17 mM	-	(Fang et al., 2014)
				DPV	62 nM	0.2 µM–0.1 mM		
			TiO_2	CV	126 nM	0.6 µM–0.17 mM		
				DPV	35 nM	0.2 µM–0.1 mM		

	Lateral flow biosensor	*Phytophthora infestans*	GNP	Color intensity (CI)	0.1 pg. μL^{-1}	0.1-100 pg.μL^{-1}	1.5 h	(Zhan et al., 2018)
	Gold electrode	*Penicillium sclerotigenum*	MNP	EIS	-	50 -200 pg.μL^{-1}	30 min	(Silva et al., 2013)
Fungi	SPR	*Pseudocerocospora fijiensis*	-	SPR sensogram	11.7 $\mu g.mL^{-1}$	39.1 - 122 $\mu g.mL^{-1}$	25 min	(Luna-Moreno et al., 2019)
	SPR	*Phakopsora Pachyrhizi*	Gold disk	SPR sensogram	800 $ng.mL^{-1}$	3.5 - 28.0 $gm.L^{-1}$	2 h	(Mendes et al., 2009)

13

14 **Table 4.** Different biosensors proposed for bacteria pathogen detection

Method	Sensing platform	Signal mode	Nano material	Target	LOD	Working range	Analysis time	Reference
Optical biosensor	SPR	SPR sensogram	Ag-BaTiO3-graphene	*Pseudomonas* spp.	7.09 log CFU mL^{-1}	-	-	(Mudgal et al., 2020)
Colorimetric biosensor	GNP–probe DNA	UV absorbance spectrum	GNP	*P. syringae*	15 $ng.\mu L^{-1}$	2-200 $ng.\mu L^{-1}$	1 h	(Vaseghi et al., 2013)
Nanobiosensor	GNP–probe DNA	SPR band	GNP	*R. solanacearum*	7.5 ng	7.5-100 ng	15 min	(Khaledian et al., 2017)
Colorimetric biosensor	Colorimetric Probe	UV-vis spectroscopy	GNP	*R. solanacearum*	0.2 ppm	0.1-1.0 ppm	24 min	(Aoko et al., 2021)
Immunochromatic	Sensor strip	digital colorimetric analysis	-	*X. fastidiosa*	0.8×10^8 cells. mL^{-1}	$(0.8\text{-}4.1) \times 10^8$ cells. mL^{-1}	4 h	(Wen et al., 2017)
Lateral flow assay	GNP–probe DNA	CI	GNP	*Dickeya. solani*	14,000 $CFU.g^{-1}$	$0\text{-}10^8$ $CFU.g^{-1}$	30 min	(Ivanov et al., 2020)
Lab-on-chip device	gold interdigitated electrode	EIS	-	*Pectobacterium atrosepticum*	10^4 CFU. mL^{-1}	$10^4\text{-}10^9$ CFU. mL^{-1}	1 h	(Hashemi Tameh et al., 2020)

Fluorescence biosensors.	Optical system	CI	-	*C. Liberibacter asiaticus*	1.0 x 10^{-4} ng.μL^{-1}	-	40 min	(Wu et al., 2021)
Chemiresistive biosensor	SWNT-based device	Resistance changes	SWNT	*C. Liberibacter asiaticus*	5 nM	3 nM-2.6 μM	30 min	(Tran et al., 2020)

15

16 **Table 5.** Different biosensors proposed for virus detection

Method	Sensing platform	Signal mode	Nano material	Target	LOD	Working range	Analysis time	Reference
Electrochemical biosensor	SPCE	EIS, CV	GNP	*Citrus tristeza virus*	100 nM	0.1 - 10 μM	1 h	(Khater et al., 2019)
Electrochemical immunosensor	Gold electrode	EIS, CV, DPV	-	*Fig mosaic virus*	0.03 nM	0.1 nM - 1 μM	-	(Haji-Hashemi et al., 2019)
Electrochemical immunosensor	Gold electrode	EIS, CV, DPV	-	*Citrus tristeza virus*	.0.27 nM	1 nM - 5 μM	1.5 h	(Haji-Hashemi et al., 2017)
Amperometric immunoassay	Carbon electrode	CV, UV–Vis spectra	GNP	*Citrus tristeza virus*	0.3 fg.mL^{-1}	1.95–10.0 × 103 fg.mL^{-1}	1 h	(Freitas et al., 2019)
Bioelectronic biosensor	Gold gate electrode	CV	-	*Plum Pox Virus*	180 pg.mL^{-1}	5 ng.mL^{-1}- 50 μg.mL^{-1}	-	(Berto et al., 2019)

Immunological technique	Lateral flow immunoassay	-	MNP , GNP	*Potato virus Y*	0.25 ng.mL^{-1}	0-125 ng.mL^{-1}	25 min	(Razo et al., 2018)
Electrochemical DNA sensor	SPGE	CV	-	*Cucumber Mosaic*	-	1-100 ng.µL^{-1}	2 h	(Zulkifli et al., 2016)
Immunochromatography	Lateral flow immunoassay	CI	GNP	*potato virus X*	17 pg.mL^{-1}	0-0.5 ng.mL^{-1}	12 min	(Panferov, Safenkova, Zherdev, et al., 2018)
Amperometric biosensor	Pt electrode	Current signal	-	*Tobacco mosaic virus*	0.1 mM	0.1–7.4 mM	1 h	(Bäcker et al., 2017)
Immunosensor technique	SPCE	Potential signal	GNP	*cucumber mosaic tomato yellow leaf curl virus*	-	0.1-1.3 mg.mL^{-1}	-	(Uda et al., 2017)
Plasmon resonance biosensing	Colorimetric assay	UV-Vis spectra	GNP		-	1- 5 pM.µL^{-1}	-	(Razmi et al., 2019)
Lateral flow strips	Lateral flow strips	CI	-	*tomato spotted wilt*	-	-	10 min	(Lee et al., 2021)
Nanobiosensor	Fluorescence resonance	Fluorescence emission	QD	*Citrus Tristeza Virus*	198 ng.mL^{-1}	-	-	(Safarnejad et al., 2017)

	energy transfer (FRET)							
Nanobiosensor	FRET	Fluorescen -ce emission	GNP/Q D	*Citrus Tristeza virus*	130 $ng.mL^{-1}$	0 - 1 $\mu g.mL^{-1}$	1 h	(Shojaei et al., 2016)
Immunosensor	Optical biosensor	UV-Vis spectra	ZnO films	*Grapevi ne virus A-type*	-	1 $pg.mL^{-1}$ - 10 $ng.mL^{-1}$	-	(Tereshchen ko et al., 2017)
Immunoassay	Lateral flow	CI	GNP	*potato leafroll virus*	0.2 $ng.mL^{-1}$	0.2-100 $ng.mL^{-1}$	15 min	(Panferov, Safenkova, Byzova, et al., 2018)

17

4.3. E-eye (optical sensors)

A large variety of optically based sensors has been designed specifically for plant disease monitoring and reviewed over the last decade (Sankaran et al., 2010) (Li et al., 2014), (Fang & Ramasamy, 2015) (Barbedo, 2016) (Galatus et al., 2020) (Singh et al., 2020) (Mishra et al., 2020).

Techniques can be classified in many ways, for example local optical probes offer a precise measurement with limited impact of external parameters during acquisition. Imaging methods enable parallelization of the acquisition which is useful to increase the field of view ranging from proxi-detection (mm to m) to tele-detection (cm to km). While time processes in plant-disease interactions are relatively long (from days to weeks for full process of infection), the movement of the plants may cause some acquisition artifacts depending on the acquisition time of the optical sensor. Some methods are designed for *in vitro* analysis of the development of disease on leaves or biological medium while others are suitable for *in situ* monitoring. For *in situ* monitoring constraints differ whether ones operate in controlled environment or in the field. Controlled environments enable a biological control of the entire plant-pathogens interaction and a control on the lighting conditions during acquisition (Kuska & Mahlein, 2018). Field environment monitoring, while more realistic for agricultural applications is more difficult to tackle due to the possibility of multiple stresses and the non-controlled environment for optical acquisition. Other discriminant parameters include the distinction between active and passive optical systems depending on whether or not purposely chosen light is required to shine onto the plants. For active imaging systems the light sent can activate some physiological processes or just be used to generate contrast and, in this case, it is important to check that this light is in actinic, i.e. does not cause perturbation on the plant-disease interaction (Khater et al., 2019). Finally, some methods may provide features on the symptoms of the disease, defense and resistance of the plant depending on the timing of the image acquisition in the plant-pathogens interaction cycle. Fig 3 shows a schematic of optical sensor for plant monitoring.

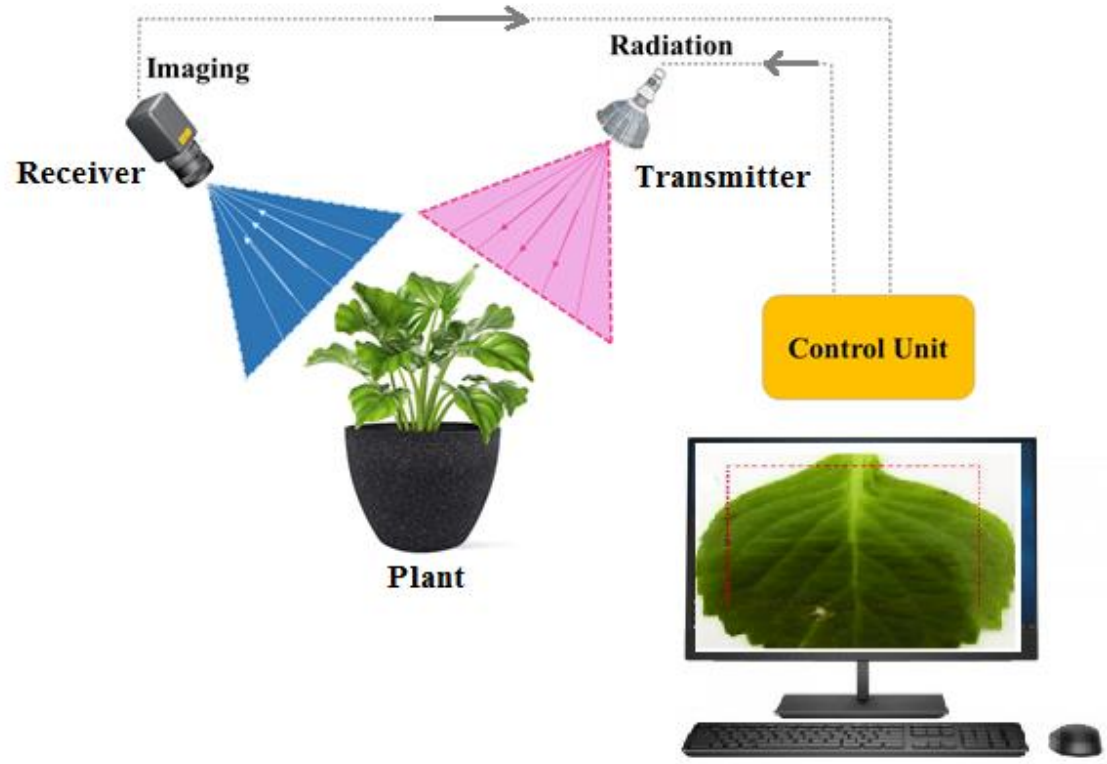

Fig 3. Schematic of optical sensing for plant monitoring

The observed contrast can originate from various physical phenomenon. Fungi sporulation, lesion, depigmentation, necrosis can create or destroy micro layers. This affects the refraction index, the absorption, and the scattering properties of the leaves. This produces contrast in reflectance in various wavelength as reported in (Barbedo, 2016; Fang & Ramasamy, 2015; Li et al., 2014; Mishra et al., 2020; Sankaran et al., 2010; Singh et al., 2020). for a large variety of plant pathogens interactions. These contrast in the spectral signature can be used in various ways: By selecting optimal wavelength and design optimal multispectral imaging or keep the entire spectrum for hyperspectral imaging. Multi and hyperspectral imaging are by far the most promising and already currently used techniques for plant-disease monitoring with optical-based sensors (Bock et al., 2010; Golhani et al., 2018; Lowe et al., 2017; Mahlein et al., 2017; Moghadam et al., 2017). Trade-offs can be considered depending on the sensitivity of the resulting optical systems, their hardware cost, weight, size, or computational load. A current trend is on the reduction of the cost of multi and hyperspectral imaging either by developing snapshot hyperspectral systems (Douarre et al., 2021; Douarre et al., 2020) or through the development of custom Bayer filters (Tisserand, 2019).

When perceptible by the human eye these contrasts can be compared to an expert ground truth. This has opened the way to the possibility of automated recognition, detection or quantification of plant diseases with supervised machine learning (Li et al., 2021). The bottleneck in the design of such algorithms now becomes the annotation of images. Indeed, after capturing images experts must watch each individual image and label them. This corresponds to a global label for recognition, bounding boxes for detection of lesion, and at a pixel level for segmentation. This error prone and time-consuming task is currently a limiting factor to the dissemination of computer vision technologies for plant disease analysis with optical systems. A method to circumvent this bottleneck is to use embedded eye tracking system capable of recording simultaneously images and the position of the human attention in these images (Samiei et al., 2020).

When gazing outside the visible spectrum, it is more difficult to establish a ground truth and the recorded contrasts must be confronted to other types of measurements to validate the physiological interpretation of the contrast. This concerns a variety of optical techniques such as X-Ray imaging, fluorescence imaging, speckle imaging, thermal imaging, terahertz imaging and magnetic resonance imaging. Most of these techniques for plant health monitoring are however of limited applicability due to the duration of acquisition time, the current cost of instrumentation or the non-transferability to field experiments.

The above criteria have been summarized in Table 6 to position the most promising optical systems found in the literature for plant disease monitoring based on review articles.

Table 6. Imaging optical-sensors for plant-disease monitoring

Technology used	Environment	Acquisition time	Application	Review Ref
X-Ray	Controlled	min	Microstructure	(Du et al., 2019)
Multi-Hyperspectral lmaging	All	s	Reflectance	(Bock et al., 2010; Golhani et al., 2018; Lowe et al., 2017; Mahlein et al., 2017; Moghadam et al., 2017)

Chlorophyll Fluorescence	Controlled	min	Photosynthesis	(Daley, 1995; Murchie & Lawson, 2013; Pérez-Bueno et al., 2019)
Speckle	Controlled	min	Microdeformation	(Zdunek et al., 2014)
Thermal Imaging	Controlled	s	Evaporation	(Ishimwe et al., 2014)
TeraHertz Imaging	All	s	Water content	(Afsah-Hejri et al., 2020)
Magnetic Resonance Imaging	Controlled	Min	Water content	(Faust et al., 1997)

4.4. Acoustic sensors

Sound, or acoustic energy, may produce in the form of an oscillating concussive pressure wave and transmitted through gases, liquids and solids. The lowest frequency in the acoustic spectrum is related to infrasound in the frequency range up to 20 Hz. Ultrasound is defined as acoustic waves at frequencies above 20 kHz, which are widely used in medical practice as a diagnostic and therapeutic tool. Ultrasound and infrasound can communicate with biological tissues through thermal and mechanical processes. Humans can detect sounds in a frequency range from about 20 Hz to 20 kHz (Speaks, 2017). Fig 4 shows principle of an acoustic-based sensor for plant disease application.

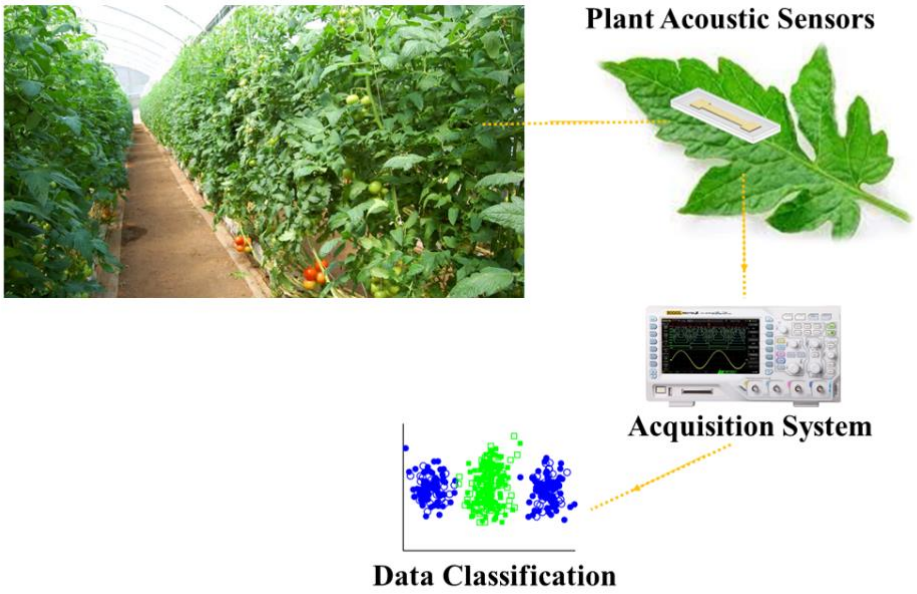

Fig 4. Schematic showing the use of acoustic sensors for plant monitoring

The physiological effect of environmental factors, such as humidity, light, wind and temperature on plant stimulus and growth are well understood (Hassanien et al., 2014). However, little information is available on the effects of sound on plants. Sound stimulation can increase resistance to disease and reduce the need for chemical fertilizers and chemicals. Plants can spontaneously generate sound waves with relatively low frequencies of 50-120 Hz. Like humans and other animals, plants may have internal preferred frequencies of vibration. Plants can also absorb and amplify specific external sound frequencies.

Dostál et al. (2016) detected the acoustic emission (AE) characteristics of the plant according to water stress conditions (Dostál et al., 2016). They concluded that the occurrence of AE signals caused by the transpiration system is related to the cavitation event in the plant. The results obtained using multiple regression analysis showed that the change in the amount of AE signal detected from the plant studied was affected the most and the least by temperature and light intensity, respectively. Using SAW (surface acoustic waves), Lee et al. (2018) captured consecutive images of increased water transfer in *Epipremnum aureum* at three different frequencies (10, 15 and 20 MHz) (Lee et al., 2018). A dye solution used at 15 MHz SAW showed the highest intensity value after 40 min of stimulation via SAW. The excitation areas for 15 and 20 MHz SAWs were respectively reduced to 42.3% and 22.6% in comparison to that of 10 MHz SAW.

Overall, the results obtained showed that regardless of excitation area, the transport of water in the leaves was maximized at 15 MHz SAW. You et al. (2011) developed an AE-based system to control crop disease stress (You et al., 2011). The results obtained showed this system is capable of adjusting to delivery volume, controlling valve speed under signals in AE and environmental information for partial crop growth, allowing the use of intelligent control to spray the crop and accordingly, reduce the consumption amount of pesticide.

Qi et al., (2009) investigated the effect of sound wave stimulation on strawberry growth in a sunlit greenhouse (Qi et al., 2009). The experimental results of this study show that sound waves not only can improve the strawberry growth, but are also capable of increasing the resistance to disease. Three reasons were proposed as to how the use of sound waves improves the plant growth: (i) the fluidity and permeability of the plant membrane undergo change due to environmental stress (e.g., sound wave stimulation); (ii) stress is signaled to other molecules by the signaling molecule Ca^{2+}; (iii) Related gene expression occurs due to the spread of stress signal. On the other hand, signal resonance will take place when the frequency between external vibration and the plants natural (spontaneous) sound are congruent.

Kim et al. (2015) showed that sound vibrations (SVs) play an important role in increasing the postharvest shelf-life of tomatoes (Kim et al., 2019). They showed that fruit treated with sound vibrations at 1000 Hz delay the ripening of tomatoes in comparison to the control sample.

Appel and Cocroft (2014) reported better defense of *Arabidopsis* rosettes against a subsequent attack by SVs after being pre-exposed to this pathogen caused by feeding of the *Pieris rapae* (L.) caterpillar(Appel & Cocroft, 2014). In comparison to untreated plants, treated plants exhibited higher levels of defence to glucosinolate and anthocyanin. Results from this study proved that SVs sensors are environmentally favorable for plants.

Using AE technology, Yang et al. (2014) developed a system for diagnosis of crop disease stress conditions (Yang et al., 2014). The results of their study showed that the continuous detection of AE signals of a disease stressed plant and according to the specific rules of the physiological cycle, the AE phenomena for the infected plants are dissimilar healthy plants.

4.5. Wearable sensors

The development of wearable sensors allows a nanotechnology-based platform as a non-invasive tool for plants. Although these sensors are expensive, the cost of these sensors should

significantly reduce over time (Guk et al., 2019; Heikenfeld et al., 2018). The use of such sensors in plant science is a useful tool in the production of agricultural products that can collect real time information about the plant physiological status. A wearable plant sensor can be easily attached onto the plant surface due to its excellent mechanical compatibility (F. Zhao et al., 2020). Sensor networks based on flexible wearable nanoelectronic circuits implanted on plants provide wireless communication via low concentrations of volatile organic molecules in real time. Fig 5 shows a schematic of plant wearable sensor for E-monitoring.

Fig 5. A plant wearable sensor for E-monitoring

The high sensitivity of the integrated arrays of SWCNT channels and graphite electrodes transferred onto the surface of the live plants leaves can track chemicals in the air. The elastic properties of nanomaterials-based wearable sensors allow them to act like a flexible skin and bend over objects with a radius of curvature below 100 μm (Giraldo et al., 2019). SWCNT-graphite-based wearable sensors that work with radio frequency (RF) can be used for wireless monitoring in combination with electronic devices without power consumption for gas molecules up to 5 ppm (Lee et al., 2014).

Carbon-nanotube-based wearable sensors for measuring plant VOCs, such as polyethylene, are commercially available for agricultural use, but still are not used for electronic monitoring. Though many graphene and carbon nanotubes- based wearable stretchable sensors have been reported for wireless control of a wide range of gas and aqueous phase molecules, including glucose (Bandodkar et al., 2016), few publications have so far reported their applications in plants.

Lan et al. (2020) developed a wearable humidity sensor based on laser-induced graphene to monitor transpiration of plant leaves (Lan et al., 2020). This wearable sensor with its high flexibility is capable of on-site monitoring of plant transpiration on the surface of plant leaves without causing prolonged and highly stable physical damage. Using this type of sensor combined with nanoparticles, such as graphene and laser technology is a promising choice for the next generation of wearable sensors to be used in intelligent agriculture. Little research has been published on the use of wearable sensors for diagnosis and management of plant diseases. A summary is given in Table 7.

Table 7. Application of wearable-based sensors for plant monitoring

Technology used	Plant	Signal mode	Nano material	Application	References
Eectrochemical biosensor	Leaf of spinach and the fruit of apple	Square wave voltammetry (SWV), CV	GNP, grapheme	Pesticide sensing/ methyl parathion	(Zhao et al., 2020)
Laser	-	Capacitance signal	Graphene	Transpiration monitoring	(Lan et al., 2020)

Microneedle patch-based sensor	Tomato leaves	UV Absorption spectra	-	Pathogen/ *P. infestans*	(Paul et al., 2019)
Electrochemical sensor	Maize plant	CV, EIS	Pt Nanoparticles	Methanol emission	(Moru et al., 2020)
OCT	Apple, pear, and persimmon Leaves	OCT signal intensity	-	monitoring quality of leaf	(Wijesinghe et al., 2017)
Smartphone-based	Tomato leaves	GC–MS spectra and Chemometric Analysis	GNP	*P. infestans*	(Li et al., 2019)
Fluorescent fiber	-	Absorption spectrum	-	Growth rate Monitoring	(Galatus et al., 2020)
Strain sensor	Eggplant	Change of resistance	CNT	Plant growth monitoring	(Tang et al., 2019)
IOT-based sensor	Rice	AgriTalk data	-	Blast Detection	(Chen et al., 2019)
Lab-on-chip device	Olive trees	EIS	-	*Xylella fastidiosa* detection	(Chiriacò et al., 2018)
Optical sensor	A leaf	Light Absorption spectra	ZIS nanosheets	Plant growth monitoring	(Lu et al., 2020)
Wireless gas sensors	Leaves of a live plant	Radio frequency	Graphene	Dimethyl methylphosphonate	(HyungáCheong et al., 2016)
Chemi-resistive sensor array	Tomato leaves	-	Graphene	Monitoring of late blight	(Li, 2021)

Tang et al. (2017) fabricated a chitosan-based flexible and stretchable sensor to capture the plant response to mechanical damage, methyl parathion, and nitrite (Tang et al., 2017). The resistance change caused by the change in strain due to mechanical damage confirmed that a wearable sensor can control plant wounding.

Oren et al. (2017) developed a flexible microscale plant sensor with graphene nanomaterial-based patterning onto different types of tapes (Oren et al., 2017). This sensor estimates the hydraulic conductivity by measuring the time taken for water transfer from root to leaf and the transpiration level of plants based on changes in the electrical resistance of graphene in different humidity environments. Hydraulic conductivity and transpiration rate of plants may

be used as an indicator for ambient temperature and drought stress in plants. Kim et al. (2019) vapor printed polymer electrodes directly on living plant tissue (Kim et al., 2019). The results of their study showed that the sensor used is capable of detecting many biological and abiotic stress factors in plants. In addition, this new electrode showed excellent adhesion to plant tissues during a 130- day period without affecting the plant's biocompatibility and natural growth pattern.

Microneedles are another type of wearable sensors, which have shown their capability in reaching vasculature inside the plants with the minimum rate of invasion. These sensors are both capable of detecting sap flow rate and extracting sap and analyzing its physicochemical properties such as pH and electric conductivity. Baek et al. (2018) manufactured a microneedle sap flow sensor on the basis of the modified Granier method for monitoring water transport in the stem of tomato plants (Baek et al., 2018). This wearable sensor can potentially be used to show the environmental variables changes, such as intensity of solar irradiance, moisture, and soil water content (SWC).

Jiao et al. (2019) developed a microneedle to detect the plant's nitrate using an insertable silicon chip consisting of a nitrate-selective field effect transistor (Jiao et al., 2019). Their wearable sensor indicated that the nitrate concentration inside the plant can be continuously detected under different light and irrigation patterns. Though thin-film wearable sensors allow for fast prefabrication and large-scale fabrication, they also have some limitations. They may lead to low accuracy and impaired normal plant function when adhering to irregular parts and complex plant tissue. This limitation will be significantly worsened due to the significant increase in the size of the plant tissue during growth. This challenge severely prevents long-term monitoring, especially for fast-growing plants such as vegetables. One way to overcome this limitation is to develop the flexible wearable sensors with greater deformation capability and good adhesion to irregular surfaces. The direct fabrication of such wearables on plant living tissues can potentially overcome this incompatible challenge because the device components can be placed along irregular surfaces (Wong et al., 2017).

The materials printing/ writing/ deposition on plant tissues under mild conditions to develop a plant wearable sensor has received lots of attention. Among desirable properties of wearables, especially when they are used on fruits, vegetables, and agricultural products are their nontoxicity and controlled degradability. Portable devices to directly print/write/deposit on plants should be developed to be used on large-scale in-field operation (Huang et al., 2020).

Plant wearables on cellular scale that are capable of detecting localized signals may lose distal signals or signals via other pathways. They also require significant construction skills at the microscale level to be successfully fabricated onto living tissues.

Microneedle-based wearable sensors do not suffer from mismatching on the plant irregular surface unless they have used an array of microneedles. However, depending on the size of the microneedle and the plant size, the invasive detection of microneedles may cause damage to the plant. By using biocompatible materials (e.g., pectin, cellulose, hemicellulose, lignin, etc.), decreasing dimensions of the microneedles, and applying other potential techniques (e.g., degradable microneedles), wounding effects such as callus formation may be minimized (Wong et al., 2017).

In summary, plant wearables propose a robust and convenient way to follow the trace of the plant growth and health-related biometric information in situ. Moreover, combination of such plant wearables with sensors that detect environmental conditions such as temperature, light intensity, and RH may provide a robust tool to study the plant-environment interface, to preserve optimum environments for plant growth, and to strengthen crop yield with minimum agricultural inputs. Drawbacks are also observed in reported plant wearables, and challenges are ahead of their broad use in agriculture. In order to cope with the drawbacks discussed above the interdisciplinary combination of material characteristics (e.g., strength, compliance, adhesion, biocompatibility, and degradability), fabrication methods (e.g., low cost, large scale), and especially the plant morphology and physiology (e.g., signaling in growth and responses to biotic and abiotic stresses, wounding, etc) should be used (Yin et al., 2021).

5. Conclusions, challenges, and perspectives

In this review paper, we have investigated and reviewed the latest e-monitoring methods, including biosensors, e-noses, wearable technology, acoustic sensors, and light-based machine vision to detect various plant pathogens, including viruses, bacteria, and fungi from laboratory to farm. In the case of optically based machine vision, much effort has been directed towards spectral imaging. In controlled environments techniques using enhanced spectral contrast have demonstrated their value to identify lesion and pre symptoms due to diseases. However, the published literature shows that these gains tend to vanish when moving to less controlled environments. This is due to the heterogeneity of lighting and the complexity of the canopy. For high-throughput monitoring of diseases with light based machine vision we believe that

increasing the spatial resolution is a more promising methodology than increasing the spectral sensitivity. Diseases have textural spatial signature that can be discriminated more easily when the spatial resolution is high.

Considering the rapid advances in nanotechnology and nano-based fabrication methods in the last decade, great advances are currently emerging in various types of biosensors, wearable sensors, bioelectronic noses, and nanostructured substrates for the analysis of plant diseases. One of the most important immediate effects of these new electronic diagnostic tools is that the accurate diagnosis of plant disease is now available to farm workers or farmers at reasonable cost. With the development of flexible tools that have biodiversity compatibility with plants, diagnostic tools are expected to be used in the form of an integrated and multi-mode detection mechanisms for rapid detection of infections caused by plant pathogens, as well as biotic stresses and plant growth control in real conditions.

Future approaches to nanotechnology-based wearable sensors will require high sensitivity and increased signal-to-noise ratio under variable environmental conditions. Relative humidity (RH) near the plant surface leads to increased sensor noise and recovery time. Therefore, wearable sensor performance depends on the type of plant substrate they are interfaced to. Using wearable carbon nanomaterial-based sensors, the optimum choice can be balanced against variable environmental conditions under different factors, such as temperature, humidity, and wind.

However, these tools used for e-monitoring of plant diseases are still accompanied by challenges, such as the environmental and toxic effects of nanomaterials used in measuring tools, data sharing speed and rapid disease prediction, and long-term sustainability of the sensor in different weather conditions, including hot and cold weather, moisture, strong sunlight, wind, heavy wear. The use of wearable sensors in the form of lab-on-a-chip as a flexible tool and compatible with plant conditions can to a large extent open a new path for e-monitoring without adversely affecting plant growth or crop production. However, currently its usage to understand some of the plant's physiological responses, such as water, nutrients and light has limitations.

Despite the challenges, the recent development of small, portable, and cost-effective tools for the rapid and accurate detection of plant diseases, plant health management and monitoring long-term, is showing great potential. The future of e-monitoring methods for developing of digital farm and precision agricultural is very promising.

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Conflict of interest

The authors declare no conflict of interest.

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