



Application of electronic nose as a non-invasive technique for odor fingerprinting and detection of bacterial foodborne pathogens: a review

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Abstract Food safety issues across the global food supply chain have become paramount in promoting public health safety and commercial success of global food industries. As food regulations and consumer expectations continue to advance around the world, notwithstanding the latest technology, detection tools, regulations and consumer education on food safety and quality, there is still an upsurge of foodborne disease outbreaks across the globe. The development of the Electronic nose as a noninvasive technique suitable for detecting volatile compounds have been applied for food safety and quality analysis. Application of E-nose for pathogen detection has been successful and superior to conventional methods. E-nose offers a method that is noninvasive, fast and requires little or no sample preparation, thus making it ideal for use as an online monitoring tool. This manuscript presents an in-depth review of the application of electronic nose (E-nose) for food safety, with emphasis on classification and detection of foodborne pathogens. We summarise recent data and publications on foodborne pathogen detection (2006–2018) and by E-nose together with their methodologies and pattern recognition tools employed. E-nose instrumentation, sensing technologies and pattern recognition models are also summarised and future trends and challenges, as well as research perspectives, are discussed.

Keywords Sensors · Pattern recognition · Foodborne pathogens · Volatile organic compounds (VOCs) · Electronic nose

Introduction

Food pathogens characterise a special form of microbial pathogens, which are acquired and spread through food. Foodborne Pathogens (*Campylobacter*, *Clostridium botulinum*, *Escherichia coli* O157: H7, *Listeria monocytogenes*, *Norovirus*, *Salmonella*, *Staphylococcus aureus*, *Shigella*, *Toxoplasma gondii*, *Vibrio vulnificus*) are a significant source of foodborne illnesses, hospitalization and deaths in the world (Havelaar et al. 2015).

The global incidence of foodborne related diseases is on the rise with a reported 600 million illnesses and 420,000 deaths every year, leading to the loss of 33 million healthy life years measured in Disability-Adjusted Life Years-DALY's (Franz et al. 2018).

Foodborne illnesses usually occur through the contamination of surfaces, oral-faecal route and improper food storage (Nygren et al. 2013). The detection of foodborne pathogens is a critical component in the elimination of pathogens in the food supply chain. Current detection methods include conventional cell culture standards, immunological assays, DNA based methods, Biosensor based methods, as well as emerging spectroscopic methods and spectral imaging techniques.

Recent developments in sensor technologies have led to innovative analytical approaches such as the electronic nose (E-nose) that been developed and applied in the food industry in reaction to emerging food safety issues. E-nose provides a rapid, non-invasive online monitoring tool for

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food safety and can be used for qualitative and semi-quantitative detection (Chen et al. 2013).

An E-nose is a device capable of identifying simple or complex odours by combining a chemical sensor array system with partial specificity and a suitable pattern recognition system (Gardner and Bartlett 1994). E-nose can analyse volatile organic compounds (VOCs) produced by microorganisms is employed as a possible alternative method in the identification and classification of different chemicals and bacteria.

E-nose has gained widespread application in the food industry and has been applied for the detection of food spoilage bacteria (Pattarapon et al. 2018; Wang et al. 2012) and total volatile basic nitrogen (Li et al. 2016), trimethylamine (Ampuero et al. 2002), fungal infections (Lippolis et al. 2018; Liu et al. 2018; Pallottino et al. 2012). E-nose presents numerous advantages over conventional and other non-invasive methods such as vibrational spectroscopic methods and hyperspectral imaging.

In this article, the following specific objectives are discussed:

1. The use of electrochemical sensors for monitoring microbial growth.
2. E-nose Instrumentation, sampling and pattern recognition methods.
3. The application of E-nose will be discussed as a non-destructive analytical tool for food safety analysis (foodborne pathogen).

Methodology

Electrochemical techniques for monitoring microbial growth

Electrochemical (EC) techniques such as electrochemical impedance spectroscopy, voltammetry, potentiometry, and coulometry have made substantial contributions to the food analysis. EC techniques directly convert chemical processes that occur in a solution at the electrode/electrolyte interface into quantifiable electronic signals such as altered conductive properties (conductometric), current (amperometric), and potential or charge accumulation (potentiometric) (Niu et al. 2014).

The application of electrochemical sensors and detectors for food analysis is expanding rapidly due to their inherent sensitivity, selectivity, and speed of detection. Electrochemistry provides a noninvasive method for monitoring microbial activity as well as for monitoring electron flow within microbial communities (Martin et al. 2018). The theoretical basis of electrochemical gas sensor operation involves interactions between gaseous molecules and

sensor-coating materials. Electrochemical gas sensors will be described during the course of this article.

Electronic nose instrumentation

An electronic nose (Fig. 1) is an artificial olfaction system that comprises of units for gas/odour sampling, sensing, signal preprocessing, pattern recognition, and odour expression (Jia et al. 2018). E-nose allows for capturing volatile chemical compounds into an array of sensors through a sampling system. Signal response is generated and subsequently transmitted to a computer system for processing and pattern recognition.

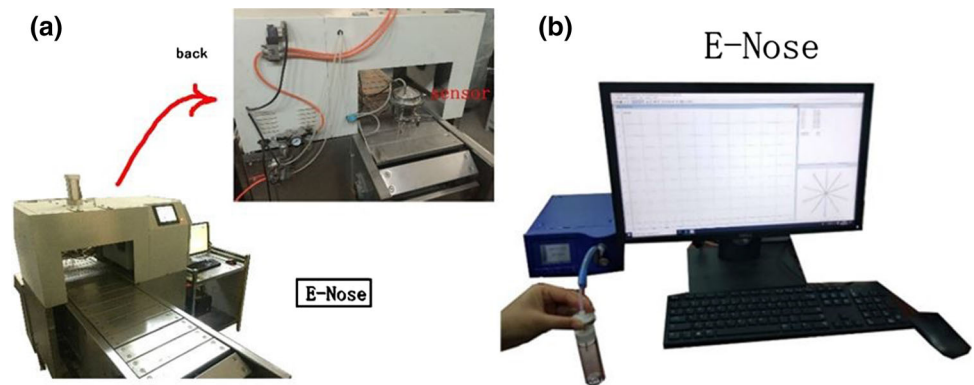
Electronic nose sensor types

Electronic noses employ an array of chemical sensors with varying specificities which reacts and respond to volatile organic compounds present in the gases collected from samples (Jiang and Chen 2014; Zohora et al. 2013). The selection of sensors to employ is quite large and have been classified into broader classes they include quartz crystal microbalance sensors, surface acoustic wave sensors, electrochemical sensors, optical sensors, and calorimetric sensors. A detailed description of these sensors together with their merits and demerits is comprehensively discussed by Wilson and Baietto (2009).

Some chemical-based sensors (catalytic, semiconducting metal oxide, solid electrolyte, polymer and field-effect transistor-based sensors) have been used for E-nose. Metal oxide semiconductors (MOS) have been commonly used as sensing elements in the electronic nose by researchers due to their availability, high sensitivity and their ability to respond to oxidising and reducing compounds. The MOS sensor is based on the adsorption of gas molecules to incite change in conductivity. The measured change in conductivity corresponds to the amount of volatile organic compounds adsorbed. One disadvantage of MOS is its susceptibility to poisoning by sulphur compounds present in the odorant mixture.

Another conductivity sensor is the polymer sensor made up of polypyrroles, thiophenes, indoles polyaniline, furan material polymers (Ghasemi-Varnamkhashti et al. 2018). Chemicals form either ionic or covalent bonds when exposed to the polymers. Changes in conductivity occur due to the transfer of electrons along the polymer chain. Polymer sensors operate at ambient temperatures and do not require heating. They are suitable for use as portable instruments having a simple electronic interface. Polymer sensors are however susceptible to humidity which can mask the responses of VOCs.

Fig. 1 **a** A fabricated E-nose machine for online detection at Jiangsu University, **b** commercial E-nose machine, Aisense PEN3 (Aisense Analytics GmbH, Schwerin, Germany)



Electronic nose sampling systems

To effectively design a pattern recognition and analysis system for electronic nose data, the processes involved in analyzing the data generated must be studied as shown in Fig. 2.

Selecting a suitable sampling of the volatile fractions and conveying it to the sensor array is a major challenge when designing the analytical methodology for microbial volatile profiling with electronic nose. The sampling technique for bacteria used usually depend on the sample state of matter (liquid, solid, semi-solid), food matrix and the level of concentration or bacterial load. Sampling systems that allow for agitation and the use of a longer sampling period to generate more volatiles are mostly preferred.

Headspace methods, analytical distillation methods or direct extraction methods are usually employed for sampling odour active analytes where adequate isolation is required. The most widely used method is the static headspace (SHS) sampling technique. It comprises of placing the microbial sample in a hermetically sealed vial after equilibrium between the matrix and the gaseous phase is established, the headspace is sampled (Peris and Escuder-Gilbert 2009). Therefore sampling techniques are

designed to be stable and be able to withstand environmental effects (Rayappan et al. 2017).

A detailed description of E-nose sampling methods is described by Majchrzak et al. (2018).

Feature extraction and dimensionality reduction

Preprocessing is the significant first step in E-nose data analysis and is usually performed to remove irrelevant information from the signal data. Preprocessing of multivariate signals is usually performed to prepare the for obtained multivariate pattern analysis. These methods are used for baseline manipulation, compression, noise reduction, detection and removal of outliers and normalisation (Sanaeifar et al. 2017).

The influence of preprocessing methods on class recognition of chemical compounds is described in several studies with Gardner et al. (1998) and Gutierrez-Osuna and Nagle (1999) using 36 and 48 different pre-processing algorithms respectively. Sensor data preprocessing methods include Scaling (dimensional auto-scaling, mean centring, relative scaling, vector auto-scaling, logarithmic scaling and power scaling), and Baseline correction (fractional difference) (Jha et al. 2019). In other to remove background noise from the raw sensor responses,

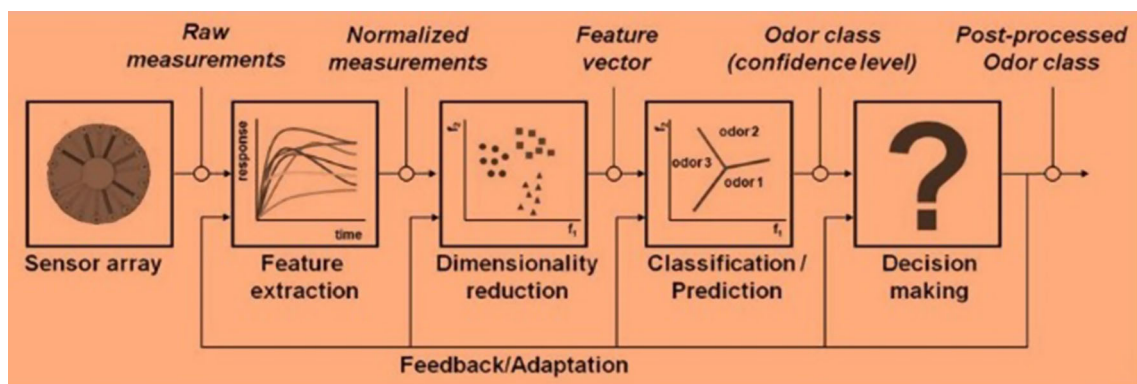


Fig. 2 Stages of classic signal processing of electronic nose data by Gutierrez-Osuna and Nagle (1999)

Fractional techniques deduct the baseline value of the sensor response value and then divide by the baseline value, to yields a per unit response. The application of fractional changes in conductance delivers the most suitable pattern-recognition performance for MOS (Hierlemann and Gutierrez-Osuna 2008).

In Vector auto-scaling, each example vector is normalized with the mean and variance computed for each example across the different dimensions whereas, in dimensional auto-scaling, the mean and variance are computed for each dimension along with all the examples in the database. The coordinates have zero mean and unit variance (Gutierrez-Osuna and Nagle 1999). In Mean centring constant terms are removed in the data so as to make the data compatible with the model. It is applied to center on the inconsistent part of the data, and to leave only the relevant variation between the samples for analysis (van den Berg et al. 2006).

Logarithmic scaling involves extracting the logarithm of previous measures. It has been applied to prevent the influence of large variations in chemical vapor concentration on sensor responses (Watson et al. 1993). Power scaling, on the other hand, applies power law for nonlinearity class separation in feature space. The raw sensor signal is scaled by suitable inverse power law to linearize the sensor output (Sunil Kumar Jha and Yadava 2011).

Autoscaling is considered the most effective preprocessing method applied to the data before feature extraction. Autoscaling involves mean centring of individual datasets and dividing it by the standard deviation for rescaling with unit variance. The main advantage is to preclude high sensor responses from dominating the analysis. The data produced by an E-nose is made up of a set of semi-independent variables from the sensor array and a set of dependent variables (Scott et al. 2006).

The general improvement of an E-nose system usually involves the optimisation of feature extraction and selection method, pattern recognition method as well as sensitive material selection and sensor array optimization for homemade E-nose devices. The latter refers to hardware selection and optimization. The primary goal of feature extraction is to extract robust information from sensor responses with less redundancy. This would ensure the overall effectiveness of the pattern recognition algorithm applied subsequently (Carmel et al. 2003).

Feature extraction methods can be grouped into three according to the source of features extraction. Firstly from curve fitting which fits the response curves based on a particular model and extracts a set of fitting parameters as the features, examples include polynomial model, exponential model, fractional function model and the S function model (Yan et al. 2015).

Secondly, from original response curves of sensors by the extraction piecemeal signal features examples include secondary derivatives, maximum values, differences, primary derivatives integrals, the adsorption slope, and the maximum adsorption slope and lastly, from applying transforms such as fast Fourier transform (FFT) and discrete wavelet transform (DWT) (Huang et al. 2006).

In addition to the above conventional feature extraction techniques, new methods have been applied in recent years. Energy vector (EV) is a vector of energy, which contains the energy of each sensor and all the mutual energies and is useful when studying the relationship between signals of sensors of the same array.

Parallel factor analysis (PARAFAC) as a multi-way data decomposition method, PARAFAC simultaneously determines the pure contributions to the dataset and optimizing each factor as a time, in trilinear systems (Zhang et al. 2014). Dynamic moments (DM) and Phase space (PS) are usually applied in dynamical systems whereas the power density spectrum (PSD) describes the distribution of power into frequency components composing that signal. The statistical average of a signal is examined in terms of its frequency content and windowed time slicing (WTS) is another recent method based on window functions. It multiplies the time response of each sensor by time windows to obtain the area values and these values are further used as features (Kaur et al. 2012; Yan et al. 2015).

Dimension reduction is achieved through principal component analysis (PCA) or independent component analysis (ICA) for uncorrelated and independent factors respectively. PCA is the most commonly used method for dimensionality reduction and feature extraction, also known as the Karhunen–Loève transform, PCA employs orthogonal transformations to eliminate colinearity in variables and the sensor array response matrix is transformed along the virtual axes of minimum correlation (Jolliffe 2014).

ICA is a linear method capable of identifying hidden factors of random variables. ICA attempts to fragment a multivariate signal into independent non-Gaussian signals (Hyvärinen et al. 2001). In ICA the sensor array response matrix is transformed along the virtual axes with minimum correlation and statistical dependency (Jha et al. 2019). Other methods employed for dimension reduction and feature extraction include wavelet transform, independent component analysis (ICA), principal kernel component analysis (KPCA) and linear discriminant analysis (LDA). In addition to the visual discrimination of chemical compounds during these processes, the resulting data is used as input for qualitative classification and quantitative estimation of chemical concentration.

Pattern recognition methods for electronic nose

Pattern recognition methods are then applied to analyse and classify the processed data and can be classified as linear or nonlinear (Fig. 3). These methods can also be classified as supervised (*k*-nearest neighbor, Linear discriminant analysis, naïve Bayes, Backpropagation artificial neural networks, adaptive resonance theory map and support vector machine) and unsupervised (*k*-means clustering, self-organizing map, fuzzy clustering and hierarchical cluster analysis). A supervised learning algorithm learns from labelled training data while unsupervised learning deals with the unlabelled data (Sizochenko et al. 2019).

Other classification categories include reinforcement learning (Reinforcement learning neural network) approaches neighborhood approaches (RMSE Neighbourhood and Similarity measure), neural networks (Feedforward neural networks, Spiking Neural Networks and Learning Vector Quantization), and decision/bagged trees algorithms.

Artificial neural network (ANN) and support vector machine (SVM) are predominantly employed for E-nose data classification due to their robustness and high accuracy. E-nose data is also known for demonstrating strong nonlinearity. Artificial Neural Networks (ANN) is another supervised learning model employed for classification and regression analysis. ANN is ambiguously inspired by the biological neural networks in the human brain and usually consist of three layers an input, output and hidden layers (Siswantoro et al. 2017).

SVM is a supervised learning model employed for classification and regression analysis. The algorithm was created by Hava Siegelmann and Vladimir Vapnik (Vapnik 2000). SVM is a powerful tool used for function estimation, nonlinear classification, and density estimation and has formed the bases for the development of kernel-based methods. A detailed theoretical discussion of SVM and its application to E-nose datasets is described (Acevedo et al. 2007; Distanto et al. 2003; El Barbri et al. 2008; Laref et al.

2018; Pardo and Sberveglieri 2005). Back Propagation (BP) learning is a method usually employed for training most of the applied ANNs with multilayer perceptron (MLP) trained by the error back-propagation algorithm the most commonly used ANN in food analysis and classification (Dębska and Guzowska-Świder 2011). A detailed overview of ANN and its application to E-nose datasets is described by (Balasubramanian et al. (2008); Luo et al. 2004). E-nose data requires training for odor discrimination or differentiation, and this is usually performed by correlating E-nose responses with chromatography (GC, HPLC), sensory analysis or calibrating with known samples (Ghasemi-Varnamkhasti et al. 2018).

Discussions

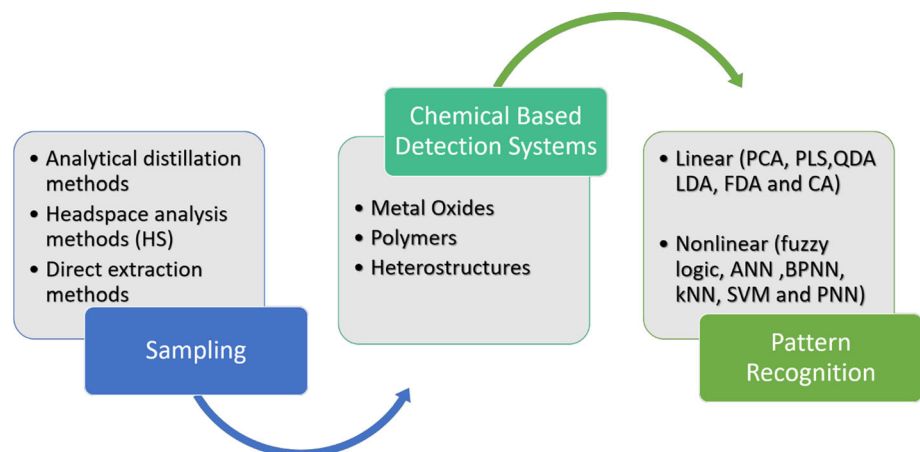
Volatiles associated with the microbial growth

Volatile organic compounds (VOCs) are a diverse group of carbon-based chemicals that are volatile at ambient temperature and can be detected through smell. VOCs have low-molecular-weights with high vapour pressures that are easily volatilized (Tait et al. 2014).

During food spoilage, pathogenic and spoilage microorganisms act upon food substrates and emit specific VOCs. This odor active molecules are generated during the process of breaking down food. Electronic nose with the requisite training program is able to discriminate amongst several volatile profiles (Giungato et al. 2018). Microorganisms have their unique characteristic volatile compounds they emit during growth (Avalos et al. 2018). Some of these volatiles provide unique odor fingerprints for a particular microorganism and can be employed for pathogen identification and discrimination without the use of conventional food analytical techniques.

In another study, Berna et al. (2013) stated that various characteristic odours are associated with pathogenic

Fig. 3 Sampling, detection and analysis of volatiles by E-nose



bacteria; *E. coli* is associated with an amino acid (indole) distinctive odour, *Salmonella typhimurium* is associated with methyl ketones, primary and secondary alcohols (Balasubramanian et al. 2016). There are no clear reasons why microorganism produces VOCs but researchers have hypothesised and attributed their production to signalling or defence mechanisms (Selim et al. 2017) as well as for growth monitoring (Kai et al. 2009).

Data of VOCs in (Table 1) reveal basic information about microbial activities at the molecular level (Robin Michael Statham and John 2012). Physiological conditions such as moisture content, oxygen, pH, and temperature

affect the composition and amount of volatiles produced by a particular microorganism. Another factor that affects the composition and amount of volatiles produced are the carbon-energy sources present for the microbes to act upon (Romoli et al. 2014). VOCs produced mainly by bacteria are produced through primary metabolism (metabolites necessary for development, growth and reproduction such as DNA, amino acids, fatty acids synthesis) and secondary metabolism (organic metabolites not involved directly in normal growth and reproduction and are intermediates of the primary metabolism). Fatty acids, acetic acid, keto acids, and amino acids act as precursors during metabolic

Table 1 A summary of volatile organic compounds produced by foodborne pathogens

Foodborne pathogen	VOCs	References
<i>Escherichia coli</i>	Indole, 1-decene (<i>E. coli O157:H7 in TSYA</i>), Dimethyl disulfide, ethanol, 2-nonanone, 2-heptanone, indole, pentyl cyclopropane (<i>E. coli in tryptone-yeast NaCl super-broth</i>), 2,5-dimethyl tetrahydrofuran, dimethyl disulfide, 2-heptanone, 2 undecanone, indole, unknown, 2-tridecane, 2,5 dimethyl pyrazine, benzaldehyde, dimethyl trisulfide, 2-nonanone, nonanal, decanal (<i>Escherichia coli O157:H7 and a nonpathogenic strain of E. coli</i>)	Siripatrawan (2008a) and Senecal et al. (2002)
<i>Listeria monocytogenes</i>	Acetaldehyde, Ethanol, Acetone, 2-Methyl-propanal, 2,3-Butanedione, 2-Butanone Acetic acid, 1-Butanol,3-Methyl-butanol,2-Methyl-butanol,3-Methyl-3-buten-1-ol 3-Hydroxy-2-butanone, Dimethyl disulfide, Pyrazine, Pyrrole, Hexanal, Butyl ester acetic acid,3-Methyl-2-butenal, Methyl-pyrazine, Methoxy-phenyl-oxime 2,5-Dimethyl-pyrazine,4,6-Dimethyl-pyrimidine, D-Limonene, 6-Methyl-5-hepten-2-one, Octanal 2-Ethyl-1-hexanol, Benzaldehyde, 2-Ethyl-6-methyl-pyrazine,2-Ethyl-5-methyl-pyrazine Pentyl-cyclopropane, Nonanal, Benzeneacetaldehyde, Acetophenone,1-Nonanol Phenylethyl alcohol, Decanal, Tetradecane,1-Ethylidene-1H-Indene,1,5-Dimethyl-naphthalene, Butylated Hydroxytoluene (<i>Tryptone soy broth</i>)	Yu et al. (2014)
<i>Salmonella</i>	Primary alcohols (1-octanol, 1-decanol), secondary alcohols (2-undecanol, 2-tridecanol), methyl ketones (2-nonanone,2-undecanone), 3-methyl-1-butanol (<i>S. typhimurium in tryptic soy yeast agar</i>), Hydrogen sulfide, ethanol, carbon disulfide, dimethyl cyclopropane, 1-propanol (<i>S. typhimurium in tryptone-yeast NaCl super-broth</i>), Dimethyl sulfide, carbon disulfide, heptane, acetic acid, ethyl acetate, methyl alcohol, ethyl benzene, 1-pentanol, 3-octanone, 3-octanol, 1-hepten-3-ol (<i>S. typhimurium in alfalfa sprouts—glass vial</i>)	Senecal et al. (2002), Siripatrawan and Harte (2007) and Siripatrawan (2008a)
<i>Staphylococcus aureus</i>	Isovaleric acid, 2-methyl butyric acid, isobutyric acid, 1-hydroxy 2-propanone, 1-hydroxy 2-butanone, butyric acid, 4-methylhexanoic acid (<i>S. aureus in blood agar</i>)	Preti et al. (2009)
<i>E. coli</i> , <i>S. sonnei</i> , <i>S. typhimurium</i> , <i>Bacillus cereus</i> , <i>L. monocytogenes</i> , <i>S. aureus</i>	1 Octanol, 1-decanol, dodecanol, 2 undecanone, 2-tridecanone, indole (<i>E. coli</i>), 1 octanol, 1-decanol, dodecanol, 2-nonanone,1-undecene, 2-undecanone, 2-tridecanone (<i>S. sonnei</i>), 1 octanol,1-decanol, dodecanol (<i>S. typhimurium</i>), 2-undecanone, dimethyl disulfide (<i>B. cereus</i>), 2-undecanone, 2-tridecanone, vdimethyl trisulfide (<i>L. monocytogenes</i>), 2-tridecanone, dimethyl disulfide (<i>S. aureus</i>)	Elgaali et al. (2002)
<i>Shigella sonnei</i>	Methanethiol, dimethyl sulfide (TSA)	Warren et al. (2007)

oxidation of glucose resulting in the production of some Microbial Volatile organic compounds (MVOCs) (Selim et al. 2017).

Bacterial volatiles (Fig. 4) is usually dominated by alcohols, furans, alkenes, aldehydes and ketones, terpenoids, sulphur compounds, acids and esters (Piechulla and Degenhardt 2014). About 346 known volatile compounds have been reported as bacterial VOCs. Reported volatile compounds classification is shown in Fig. 4 with an example of volatile compounds in each category. This volatiles may differ in species and that allow for species differentiation, although researchers for same species reported different Volatile compounds due to different substrates and detection times. Volatile compounds produced by foodborne pathogens during microbial growth in food samples are employed to characterise these pathogens.

Application of electronic nose technology for foodborne pathogen detection

As described previously, several authors have reported the detection and measurements of VOCs associated with bacterial foodborne pathogens, this had led to the attempts to create a profile of microbial VOCs (MVOCs) for a particular pathogen. The application of E-nose for pathogen detection is described in detail in this section and summarised in Table 2.

Strain and species discrimination

Green et al. (2011) successfully distinguished between *E. coli* and *Listeria innocua* in phosphate-buffered saline by employing electronic nose based on metal oxide sensor (MOS) and uncorrelated linear discriminant (ULDA) analysis with a classification accuracy of 92.4%. This method was established on odor signature processing of single bacteria colonies removed directly from the surface of the agar medium. Bacteria identification based on the E-nose response of single colonies allows for rapid results and reduces the need for culturing serological and biochemical tests.

Green et al. (2014) investigated the reliability of using E-nose for bacterial identification at the genus level using individual colonies. Four non-pathogenic bacteria species (*E. coli* DH5 α , *Listeria innocua*, *Enterococcus faecalis*, *E. coli* Biotype I) were used for this study, achieving a classification accuracy of > 80%, with a higher classification of 96.7% when E-nose sampling was repeated for the same colony and using all existing odor responses for sample characterization. Rapid identification of *L. monocytogenes*, *Staphylococcus lentus*, and *Bacillus cereus* was carried out by integrating E-nose data with chemometrics (Yongxin and Zhao 2012). Results from the study showed discrimination of four different strains of *Vibrio parahaemolyticus* with PCA explaining nearly 99% of the

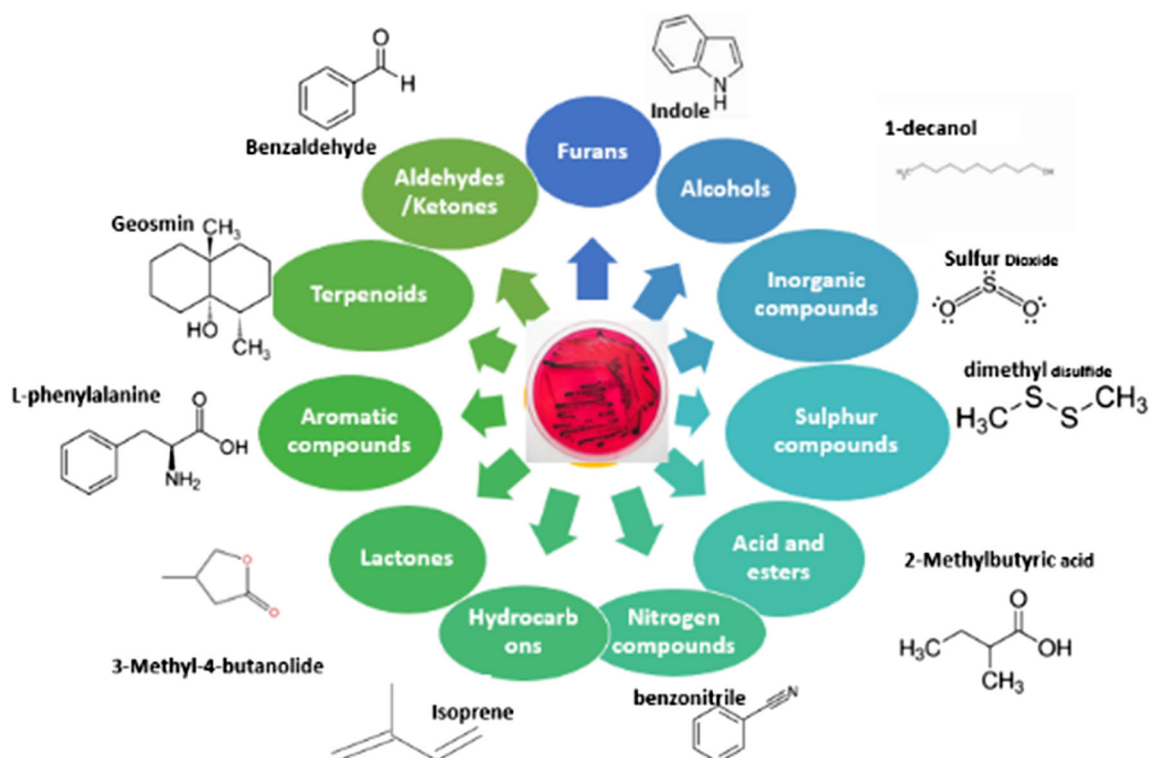


Fig. 4 Categories of chemical classes of VOCs for bacterial identification

Table 2 Recent studies on food pathogen detection by electronic nose from 2013 to 2018

Pathogens	Matrix	Sensor types	Chemometric analysis	References
<i>E. coli</i> O157:H7, <i>Salmonella typhimurium</i> 857, <i>Staphylococcus aureus</i> 29213, <i>Pseudomonas aeruginosa</i> 27853	Beef	32-polymer sensor nose chip		Abdallah et al. (2013)
<i>Salmonella typhimurium</i>	Beef	8 MOS sensors	LDA, QDA	Balasubramanian et al. (2012)
<i>Salmonella typhimurium</i>	Fresh alfalfa sprouts	12 MOS sensors	PCA	Siripatrawan and Harte (2015)
<i>E. coli</i> , <i>Salmonella typhimurium</i>	Super broth	12 MOS sensors	PCA, BPNN	Siripatrawan (2008a)
<i>E. coli</i> , <i>Listeria innocua</i> .	Lysogeny broth, Brain–Heart Infusion media	12 MOS sensors	ULDA	Green et al. (2011)
<i>E. coli</i> DH5 α , <i>Listeria innocua</i> , <i>Enterococcus faecalis</i> , <i>E. coli</i> Biotype I	Brain–heart infusion	12 MOS sensors	PCA, ULDA	Green et al. (2014)
<i>E. hormaechei</i> and <i>E. coli</i>	Mixed vegetable soups	4 MOS sensors	LDA	Gobbi et al. (2015)
<i>Escherichia coli</i>	Processed tomatoes	6 SMO sensors	PCA,	Concina et al. (2009)
<i>Staphylococcus</i> , <i>Salmonella</i> , <i>Shigella</i>	Apple	6 SMO sensors	PCA, HCA	Ezhilan et al. (2018)
<i>Escherichia coli</i> , <i>Listeria monocytogenes</i> , <i>Salmonella typhimurium</i>	Brain Heart Infusion	4 MOX thin film gas sensors and 2 MOX nanowires gas sensors	PCA	Sberveglieri et al. (2015)
<i>Listeria monocytogenes</i> , <i>Staphylococcus lentus</i> , <i>Bacillus cereus</i>	–	18 MOS sensors	PCA, HCA, DA, ANN	Yongxin and Zhao (2012)
<i>Escherichia coli</i>	Goat meat	32 polymer sensors	PCA	Ding et al. (2010)
<i>Escherichia coli</i> O157:H7, <i>Salmonella</i> spp.	Lettuce		PCA	Berna et al. (2013)
<i>L. monocytogenes</i> standard strains	Brain heart infusion broth	18 MOS sensors	PCA, HCA, ANN	Xue et al. (2012)
<i>Enterococcus faecalis</i> , <i>Escherichia coli</i> and <i>Staphylococcus aureus</i>	Street foods	9 MOS sensors	SVM	Balbin et al. (2017)
<i>Salmonella enterica</i>	Poultry manure	12 MOS sensors	ANN	Kizil et al. (2015)
<i>Salmonella typhimurium</i>	Beef	7 MOS sensors	ICA, PCA	Balasubramanian et al. (2008)
<i>Escherichia coli</i> (ATCC 25922)	Alfalfa Sprouts	12 MOS sensors	ANN	Siripatrawan et al. (2006)
<i>Escherichia coli</i> (ATCC 25922)	Packaged fresh vegetable	12 MOS sensors	SOM	Siripatrawan (2008b)

variance and discrimination of four different species of *Pseudomonas* by cluster analysis (CA) and PCA.

Xue et al. (2012) proposed E-nose together with chemometrics for strain and species-level differentiation. *L. monocytogenes* cultured on brain heart infusion broth. In this study PCA integrated with ANN for feature, extraction was successful in the identification of volatile metabolites of nine strains of *L. monocytogenes* and four species of *Listeria* spp.

These results showed E-nose has clear potential as an accurate early diagnostic screening tool for bacterial foodborne pathogen detection since discrimination between

individual bacterial colonies at both species and strain level was possible. This is crucial since virulence and pathogenicity are often associated with only a subset of bacterial strains and it is importance for a technique to have the ability to distinguish between pathogenic and non-pathogenic strains during a foodborne outbreak.

Food matrix detection

Meat products Balasubramanian et al. (2008) achieved successful prediction of *Salmonella typhimurium* in contaminated beef using E-nose data and independent

component analysis (ICA). A stepwise linear regression prediction (SLRP) model was built with the independent component (IC) and principal components (PC) with a prediction accuracy of 69.64% and 82.99%, and a root mean squared error (RMSE) of 1.358 and 0.803 for PCA and ICA respectively. The results showed that ICA performed better than PCA on the E-nose dataset, ICA which is higher-order statistical techniques can explore higher-order information of the original inputs than PCA (Cao et al. 2003).

Balasubramanian et al. (2012) compared two different gas sensor-based artificial olfactory systems i.e. conducting polymer-based and metal oxide-based sensors to successfully screen *Salmonella typhimurium* in beef. LDA and QDA classification models achieved varying levels of success for polymer E-nose (69%), metal oxide E-nose ($\geq 70\%$) and a fusion of the sensors ($> 80\%$) for classifying “No Salmonella” (microbial counts $< 0.7 \log_{10}$ cfu/g) and “Salmonella inoculated” (microbial counts $\geq 0.7 \log_{10}$ cfu/g) in meat samples stored at 10°C . The use of only relevant sensors (through Fisher Criteria Ranking of sensors) and sensor fusion approaches proved important in achieving higher classification accuracies.

Ding et al. (2010) employed a Cyranose-320 E-nose based on 32 polymer sensors for the rapid detection of *E. coli* in goat meat samples with preliminary results showing 18–77% detection accuracies for cultured bacteria. There was no differentiation between PCA data generated for contaminated and uncontaminated meat samples due to overlapping or very close marking, also the sensor was responsive to lower concentrations of the bacteria.

Street foods are a major source of foodborne illnesses, Balbin et al. (2017) applied SVM on E-nose signals for the identification and classification of *E. coli* and *Staphylococcus aureus* in street foods. Results from the study revealed successful detection of the pathogens in the street foods (Kwek–Kwek, pork barbeque and isaw) before and after cooking showing the use of E-nose as an online tool for process monitoring during food preparations.

An E-nose with a 32-sensor nose chip was applied by Abdallah et al. (2013) to detect *E. coli* O157: H7, *Salmonella typhimurium* 857, and *S. aureus* 29213 in fresh and frozen beef. Results from the study showed a strong correlation ($p < 0.005$) in gas concentration before and after the samples were contaminated with the pathogens.

Fruits and vegetables Concina et al. (2009) applied E-nose for the detection of microbial contaminants in processed tomatoes. *E. coli* with both KNN pattern recognition method showing good classification scores of 83% after 48 h from inoculation. The study revealed the influence of microorganism metabolic kinetics, on the headspace composition during microbial growth.

Siripatrawan et al. (2006) collected volatile metabolites produced by *E. coli* using an E-nose with 12 metal oxide electronic sensor. The data generated was employed to predict the *E. coli* numbers in packaged alfalfa sprouts using the ANN model with a regression coefficient (R^2) = 0.903.

Siripatrawan (2008a) developed a rapid method for differentiating *E. coli* and *Salmonella typhimurium* by combining E-nose data with PCA and BPNN models. PCA was employed for data exploration and dimensionality reduction and to successfully visualized class separation amongst sample subgroups. BPNN achieved successful prediction with a regression coefficient $R^2 = 0.96$ between true and predicted data.

A Self-organizing map () algorithm was applied for the classification of *E. coli* in packaged fresh vegetable by Siripatrawan (2008b). The SOM algorithm combined with the data from E-nose successfully classified *E. coli* above higher than 10^5 cfu/g in the vegetable samples. In a more recent study, Siripatrawan and Harte (2015) applied the Kohonen network for data visualization of *Salmonella typhimurium* present in packaged fresh alfalfa sprouts. The Kohonen network could visually distinguish different levels of *S. typhimurium* contamination on the self-organising map (SOM). The Kohonen network was valuable and better at visualizing multi-dimensional nonlinear data and showed a much more perfect separation of different sample groups than a conventional linear principal component analysis (PCA) approach.

Gobbi et al. (2015) achieved a rapid diagnosis of *E. coli* in vegetable soups. E-nose with four metal oxide sensors together with LDA analysis achieved a classification performance of 98% for *E. coli* contamination at a detection threshold of 8 and 3 cells/100 ml. The discrimination of bacterial contamination in this study was independent of the initial and final microbial concentrations. The study showed the possibility of diagnosing bacterial contamination during growth however it must be noted that the release of VOCs from bacteria changes during their growth is unknown.

Ezhilan et al. (2018), a trilayer approach, based on a homemade E-nose was used to study the presence of *Staphylococcus*, *Salmonella* and *Shigella* bacteria in delicious royal apple from the order of zero, 10^2 , 10^3 – 10^4 cfu/mL. Voltage responses for E-nose sensors together with PCA and wards HCA was applied to analyze the samples. The developed E-nose combining data classification schemes, bacterial culture study, and GC–MS analysis successfully assessed freshness or contamination levels of the apple samples.

Others Salmonella enterica is a pathogen usually associated with poultry and *S. enterica* is primarily transferred via manure contamination during processing. Kizil et al.

(2015) applied E-nose to detect the presence of *S. enterica* poultry manure with the ANN model achieving a classification accuracy of 94% for both training and validation sets. E-nose application in food quality control was investigated by Sberveglieri et al. (2015) for the detection of microorganism in water and different food matrices by employing 6 MOX gas sensors and PCA. *E. coli*, *Salmonella typhimurium* and *L. monocytogenes* at a concentration of 9×10^8 bacteria/ml.

Future trends and perspectives

Optimization of pattern recognition algorithms using metaheuristic algorithms to improve E-nose detection

The development of intelligent algorithms for pattern recognition, feature extraction, and parameters optimization is crucial for the rapid application of E-nose for routine food analysis (Luo et al. 2018). Metaheuristic optimization algorithms have the ability to resolve complex large-scale nonlinear optimization problems, and cannot be handled other analytic approaches are beginning to gain recognition in improving E-nose pattern recognition algorithms to enhance the performance of E-noses by sensor selection (Guan et al. 2014; Jiang et al. 2017; Luo et al. 2018). Heuristic methods are applied to further enhance the performance of E-noses by sensor selection and to optimize the gas sensor array as well as dimensionality reduction of the feature matrix.

Sensor development approaches

The development of reliable drift free sensors as well as investigating new material for attaining improved selectivity is crucial in achieving commercial use of E-nose in the food industry. Performance degradation of E-nose data as a result of sensor drift (variation in the sensor response in identical measurement conditions) and noise have been widely reported (Tian et al. 2018; Wijaya et al. 2017).

A Noise filtering framework based discrete wavelet transform (DWT) for handling noisy signals generated by an E-nose sensor array was developed by Wijaya et al. (2019) with significant to existing methods. The development of low-cost disposable sensors would mitigate against the decrease in the sensitivity and specificity of sensors over time. Electronic nose as a non-invasive technique provides a better alternative for the detection of complex gas mixtures, issues such as odour identification at concentrations levels higher than those of the biological counterpart as well as providing answers with regards to the concentration of a particular compound in mixtures abound. This setback in the use of E-nose is associated with

lack of adequate biomolecules to allow the system to fully mimic the biological sense of smell.

The development in the fields of genetic engineering, biotechnology and nanotechnology has led to the improved development of biomimetic electronic nose, bio-nose, b-nose, bioelectronic noses in recent years (Wasilewski et al. 2017). This new development of bioelectronics noses provides for a more precise mimicking of human smell imprints by applying highly selective and sensitive sensors.

Low-, mid-, and high-level data fusion

Applications such as multi-sensor data fusion are said to increase the probability of classification. Several researchers have applied E-nose data fusion with other non-invasive methods such as hyperspectral imaging (Liu et al. 2019), computer vision (Xu et al. 2019), electronic tongue (Banerjee et al. 2019). A fusion of electronic nose, electronic tongue, hyperspectral imaging and computer vision data at the low, intermediate, and high-level fusion models have shown to be effective and in some instances better results than single sensor models.

Potentialities of E-nose for bacterial pathogen detection

Although the application of the above mentioned future trends and perspectives for E-nose application in the food industry have been predominantly applied for food quality analysis especially for classification purposes, future prospects for applying this technology for bacterial pathogen detection remain feasible. Microorganism detection by E-nose has some drawbacks which include low sensitivity and specificity in comparison with some microbiological and molecular methods. The detection of volatile compounds is usually interfered by a complex mixture of water vapour and carbon dioxide in the background mixture (Sanaeifar et al. 2017).

Other setbacks include a high limit of detection (LOD) as Siripatrawan (2008b) reported detection limit of *E. coli* above 10^5 cfu/g with Gobbi et al. (2015) reported sensitivity as low as 3 cfu per 100 ml for *E. coli*. This setback, however, could be solved by the application enzyme substrates to liberate exogenous VOCs of foodborne bacterial pathogens to increase the diagnostic specificity of VOCs. This is achieved by modifying bacteria growth media with substrates that liberate unique VOCs through enzymatic metabolism in response to the presence of enzyme activity exhibited by a target pathogen. This methodology has been successfully studied using conventional detection methods such as gas chromatography-ion mobility spectrometry (GC-IMS) and gas chromatography-mass spectrometry (GC-MS) and can be improved with electronic nose application which is noninvasive. Example substrates

include 2-nitrophenyl- β -d-glucuronide (*E. coli*), 2-nitrophenyl- β -d-glucopyranoside (*Listeria* spp.) and 2-nitrophenyl- β -d-galactoside-6-phosphate (*Staphylococcus aureus*).

Conclusion

E-nose provides an ideal methodology for in-line process control with straight and rapid discrimination of numerous compounds requiring less or no sample preparation as well as reagent consumption. In as much as laboratory-based assessments have shown good classification rates a number of challenges with regards to humidity influence, selectivity, sensor drift, signal to noise ratio and redundancy of sensors must be resolved before the technology is moved into real-time industry application.

The influence of environmental conditions such as temperature and humidity is another drawback. Metal-oxide sensors exhibit swift response and recovery times but require high power levels to the sensors at elevated temperatures. Polymer sensors are cheap to use and operate at room temperature, however, they are sensitive to temperature and humidity.

Improved modelling and correlation between the existence of chemical markers and sensor responses together with a carefully selected sampling system and sensor arrays would greatly enhance the accuracy of results obtained from E-nose data analysis.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

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