

S. Yurish
Editor

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Chapter 7

Electronic Noses and Electronic Tongues

Tomasz Dymerski

7.1. Introduction

The human senses such as smell and taste are still main tools utilized in many areas of daily life, even the fact there were numerous attempts to elaborate a devices capable to perform their function. Nevertheless, the past 30 years of the evolution of sensor technologies gives a great opportunity to apply a newest technical solutions into the systems where sensor arrays are utilized. The electronic noses and electronic tongues are still being developed in order to measure aroma and taste in a way parallel to the biological senses. Despite of the fact these devices are not used for routine analysis, their prospective application might constitute a reasonable option too many issues where rapid analysis is advisable.

This contribution summarizes the achievements on the field of artificial senses, such as electronic nose and electronic tongue. It examines multivariate data processing methods and demonstrates a promising potential for rapid routine analysis. Main attention is focused on detailed description of sensor used, construction and principle of operation of these systems. A brief review about the progress in the field of artificial senses and future trends in concerned. A special attention has been paid to the application of these systems in two dominant fields, namely in food investigations and environmental monitoring.

Tomasz Dymerski
Department of Analytical Chemistry, Faculty of Chemistry, Gdańsk University
of Technology, Gdańsk, Poland

7.2. Senses of Smell and Taste

Gustatory sensations perceived by mammals originate when particles of volatile substances come into contact with taste receptors – specialized chemoreceptors clustered in taste buds that are located in the oral cavity. Clusters of these taste buds are located on small papillae which, depending on their location differ in shape and size. Adult humans have app. 10000 taste buds. Within each taste buds there are about 50-150 rod-shaped taste cells, which transmit the information to neuron cells which, in turn, transmits it to the brain. Five types of taste receptors which react to particular groups of chemicals present in foodstuffs or atmospheric air. Different taste sensations have different taste thresholds, highest for sweet and salty, and lowest for bitter foods. Taste sensations can be classified according to the taste-distinguishing mechanism into two groups. In the case of sour and salty tastes, the mechanism is based on hydrogen and sodium ions, respectively, reacting directly with ionic channels by changing the membrane potential of receptors [18, 23]. In the case of sweet and bitter tastes, there are protein receptor spots connected with the G protein, which, after forming a complex with a taste substance molecule, activate the G protein, leading to a series of chemical changes [4]. Both mechanisms lead to the excitation of a nerve pulse transmitted to the brain.

Sense of smell, together with the sense of taste, can be described as a ‘chemical sense’. It is the ability to discern chemical compounds or their mixtures in the surrounding air [18, 24]. The working of the sense of smell can be divided into the following steps: learning to identify and differentiate flavours, remembering and integrating different odours [25]. The ability to integrate different flavours that is exhibited by the sense of smell is a very sophisticated function which allows for the prediction and correlation between components that will combine to create a new flavour [26]. The human olfactory apparatus is able to discriminate between app. 10 000 flavour compounds with high sensitivity and accuracy [15, 22], with sensitivity in the order of parts per trillion in the case of certain chemical compounds. In some cases, even stereoisomers can be distinguished [22]. For example methyl mercaptan, a compound responsible for the characteristic aroma of garlic, has an odour threshold of less than 500 pg/L of air [21]. However, for substances such as ethane, butane or acetylene the odour detection threshold is much higher [14]. Contrary to the taste of smell, it is difficult to classify aromas into distinct groups. It has been suggested that the number of basic classes of olfactory sensations varies from 7 to 50 [22].



Molecules of odorous substances are usually volatile (molecular masses up to app. 300 Da), small, polar and often hydrophobic [13, 14] organic compounds that contain one or two functional groups [27]. Simple odours, like that of ethyl alcohol, contain only one type of odorant molecule, whilst more complex ones can consist of several thousand different chemical components, each at a different concentration level [14, 15].

The sense of smell is one of the most primeval senses from the evolutionary point of view. The part of brain in which it is located is not dissimilar to its reptilian equivalent, which suggests that it developed before mammals have emerged as a distinct class of vertebrae. Mammalian olfactory receptors (OR) are seven-transmembrane domain G protein-coupled receptors and are encoded by a large number of genes. In the case of humans, 399 intact OR genes and 297 OR pseudogenes have been identified, which amounts to app. 3 % of the entire human genome [30, 31]. The human nose is app. triangular in shape, partitioned by an internal wall called the nasal septum. It is limited from the top by an orbital lamina perpendicular to the ethmoid bone, and from the bottom by a tetragonal copula [21, 26, 34]. The nasal cavity consists of two nasal tubes separated by a partition. The olfactory epithelium is located in the upper part of the nasal fossa, in the region of the nasal septum, the roof of the nasal cavity and at the front end of the superior nasal concha [21, 24]. Olfactory cells are neurons with a dual function, serving both as chemical receptors and pulse-conducting cells [21, 26, 34]. Olfactory cells are exposed to the atmospheric air, which makes them the only nerve cells in the human organism that receive the stimulus directly from the outside world. Only about 2 % of an aromatic substance reaches the olfactory epithelium which is a mechanism that protects ORs [21, 34]. In order to stimulate a single cell, less than 10 molecules are needed. Hydrophobic molecules are dissolved in the mucosa, which increases their concentration level. After a flavour molecule becomes bound to the protein cilia of the first neuron, redundant molecules are removed through mucus efflux, enzymatic degradation in sustentacular cells, and permeation to the intercellular space and to the vascular system [4, 21, 26]. From there, information is transmitted to the brain, where the analysis of sensation features is performed, allowing for discrimination of flavours and estimation of their intensity [24, 36].

The senses of taste and smell are functionally linked [4, 36] and connected with functions of the digestive system. This is demonstrated by the fact that a temporary inhibition of the sense of smell, e.g. by



extensive mucosa production during common cold infection leads to changes in perception of a meal's flavour. The sense of smell and taste was the basis for concept of the construction of devices which mimics human senses. A certain analogy in working principle is shown in Fig. 7.1.

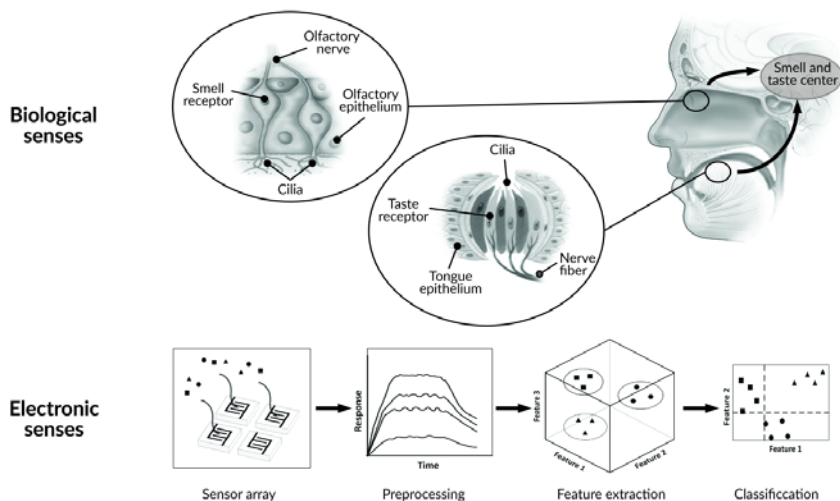


Fig. 7.1. Comparison of the sense of smell and taste with artificial senses.

7.3. Electronic Nose

An electronic nose is a device intended for the analysis of mixtures of odour substances, the operating principle of which is parallel to the mammalian olfactory sense. Devices of this type are also called odour-sensing systems [1], aroma sensors [2], electronic olfactometry [3], multisensory array technology [4], odour sensor, mechanical nose or artificial nose. At first, e-noses were equipped with arrays of sensors that could be considered analogues of olfactory receptors located in the human olfactory epithelium. Currently, devices based on different detection systems, e.g. fast gas chromatography or mass spectrometry are becoming increasingly popular [5]. For that reason, a more contemporary definition of electronic noses is an analytical device with the use of which it is possible to identify and classify mixtures of volatile chemical compounds in a relatively short time. The term ‘electronic nose’ was first proposed by Julian Gardner in 1988. However, the technique itself dates much further back. First devices of this type, albeit

quite unsophisticated [6, 7], were introduced 40 years before the term was coined. In 1961 Moncreif created a device mimicking the olfactory sense equipped with 6 different sensors [8, 9]. First work describing a multi-sensory system equipped with chemical sensors was published by Persaud and Dodd in 1982, who were able to identify 20 odorants [10]. The golden age of the development of artificial senses came in 1990's, coinciding with major developments in the construction of chemical sensors. During this decade first commercial electronic noses were introduced to the market, e.g. Alpha M.O.S. in 1993, Neotronics and Aromascan in 1994 and Bloodhound and HKR Sensorsysteme in 1995. In 1998 mass spectrometry was used for the first time in this type of devices [11]. However, chemical sensors utilized in these early e-noses had some limitations, the most common being the need for frequent calibration, lack of stability, susceptibility to poisoning, response signal masking (e.g. by ethanol) and large power consumption. With the introduction of newer types of sensors, like MOSFET or piezoelectric sensors [12], some of these drawbacks were eliminated. When using MS-based electronic noses there is no issue with profile masking, sensor poisoning, and the impact of ambient conditions (e.g. relative humidity) on the response or non-linearity of the signal. Currently, the development of electronic noses is focused on increasing the sensitivity of the device's measuring array, which would allow to detect subtle differences between similar samples, and on miniaturization, limiting the power consumption and design of portable devices.

During the analysis using an electronic nose the output signal consists of responses of individual chemical sensors which, when considered holistically, create a characteristic aroma profile of the analyzed sample. Such aroma profile is unique for each mixture of aroma compounds and is sometimes called a 'fingerprint' or 'smellprint'. The identification of odour substance is achieved by comparing its fingerprint with the response to a reference sample [4]. There are many parallels between the mode of operation of an electronic nose and olfactory sense. Chemical sensors work like olfactory cells and send signals to the pattern recognition system which, similar to brain, identifies the aroma [13, 14]. E-noses are usually comprised of an array of sensors placed in an insulated chamber, a sometimes thermostated sample chamber, electronic components (e.g. analogue-digital converter), and a pneumatic system (vacuum pump, valves, flow meters) (Fig. 7.2). For data processing software capable of statistical analysis is used.



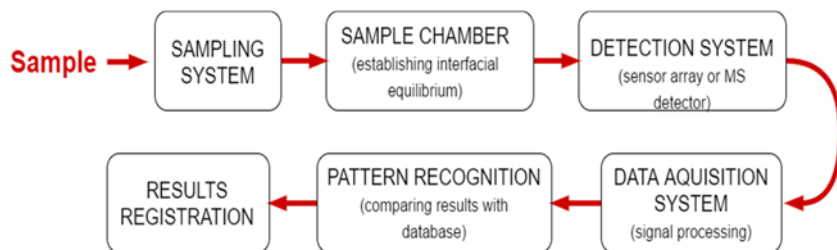


Fig. 7.2. Construction of electronic nose.

Sampling of volatile fraction can be done in a number of ways, depending on application and design of the device. Among others, direct headspace sampling, diffusion-based methods, bubbling or enrichment methods (e.g. solid-phase microextraction) are being used. A considerable challenge for constructors is the need to build each element which comes into contact with the sample out of an odourless, non-adsorptive, chemically inert materials. Both the sensor chamber and its volume should be designed in such a way, as to leave enough room to generate headspace [15]. Adequate sample incubation can be used to increase the concentration of volatile compounds in the headspace. Since the response of many types of sensors is influenced by changes in temperature and relative humidity it is necessary to monitor these parameters. Sample's headspace is carried by an inert gas or by ambient air to the sensor chamber [16]. The pump, characterized by low pulsations, is usually placed after the sensor chamber and operates in the vacuum (suction) mode, which reduces the risk of the analytes adsorbing on its elements. Before each analysis it is necessary to establish a base line of sensor's responses and measure the background noise. This is usually achieved by directing a stream of ambient air or inert gas through the sensor chamber [17, 18]. Chemical entities present in the gas come into contact with active elements of chemical sensors which leads to a change in their response signal due to, e.g. a change in conductivity. Intensity of the signal depends on the type of chemical substance, its affinity to the sensor's active element, and on its concentration. Collected response signals which form the distinct fingerprint are then processed using statistical methods. Chemometric techniques are used for data analysis (see sub-chapter 6). The comparison of the fingerprint with the odour profile of previously measured standards allows for identification and classification of the analyzed sample.



7.4. Electronic Tongue

The history of electronic tongue and nose starts in the beginning of the 20th century. Then the ion exchange theory was developed which has resulted in the construction of glass membrane electrodes used for measuring pH [19]. In the following twenty years, new sensors were designed, inter alia, MOSFET, BAW, ion exchange membrane, potentiometric biosensor, ISFET, PdMOS and SAW. In 1985, Otto and Thomas presented the first system for liquid phase analysis by using a multisensor array [20]. Seven years later, at the University of Kyusho the taste sensor was constructed by Toko; it consisted of ion-selective lipid membranes immobilized in PCV polymer [22, 23]. In 1995, as a result of cooperation between Russian and Italian research groups, a concept of electronic tongue was presented. It was based on inorganic chalcogenide glass sensor which enables both qualitative and quantitative analysis [23]. The most important facts related with development of both electronic tongues and electronic noses was listed in Table 7.1.

An electronic tongue is based on the same principles as electronic noses, with the most significant difference being that they are used to analyse solutions (mostly aqueous) instead of their volatile fractions. It is dedicated to identify, classify and analyze in a qualitative and quantitative way mixtures by applying a fingerprint method [6, 7]. An electronic tongue consists of three main elements, i.e. the sample-dispensing chamber or autosampler (optional), an array of sensors of different selectivity, and software for data processing (recognition system which mimics the brain functions) (Fig. 7.3) [30].

Application of this technical solution is consistent with the purpose of use of the human sense of smell. The array of sensors is mostly comprised of potentiometric, voltammetric, ion-selective field-effect transistor (ISFET), piezoelectric, and optical sensors. A major advantage of this type of devices is the fact, that they can be used to analyse liquid samples without any prior preparation, and solid samples after a simple dissolution [32, 33]. The sensors comprising the detection module of an electronic tongue are often not permanently fixed in an array. Because of that, the composition of the sensor matrix can be easily modified, making it flexible and adjustable to specific tasks. A unique feature of e-tongue devices is the possibility to correlate the results obtained using this technique with the results reported by a human sensory panel, which is particularly important for the food industry. They can be used to



supplement the work of trained panelists especially in a situation when it is uncertain whether the analyzed substance is safe to consume.

Table 7.1. Chronological listing of most important achievements related to the development of electronic noses and tongues.

Year	Achievement	Author(s)	Artificial sense	Ref.
1906	Relationship between pH and the potential measured in glass membrane	Cremer	e-tongue, e-nose	[19]
1909	Development of glass electrodes	Haber, Klemensiewicz	e-tongue, e-nose	[19]
1936	Commercial production of glass electrodes	Beckman	e-tongue, e-nose	[19]
1937	Nikolsky equation	Nikolsky	e-tongue, e-nose	[19]
1937	Crystalline membrane	Nikolsky	e-tongue, e-nose	[19]
1954	First gas sensor	Hartman	e-nose	[24]
1959	First MOSFET sensor	Kahng and Atalla	e-nose	[24]
1960	Semiconductor gas sensors based on metal oxide	Taguchi	e-nose	[25]
1961	First mechanical instrument for sensing odors , i.e. an array of six sensors with different coatings	Moncrief	e-nose	[26]
1964	Artificial odor-recognition system	Wilkins and Hartman	e-nose	[27]
1964	BAW sensors	King	e-nose	[27]
1965	Research on the modulation of conductivity and contact potential for the use in odor monitoring	Buck, Dravnieks and Trotter	e-nose	[28]
1967	Ion-exchange membrane	Ross	e-tongue	[19]
1969	Potentiometric biosensor	Guilbault, Montalvo	e-tongue	[19]
1970	Ion-Sensitive Field Effect Transistor (ISFET)	Bergveld	e-tongue	[19]
1973	Invention of a PdMOS sensor	Lundström	e-nose	[27]
1979	First gas sensor based on a SAW oscillator	Wohltjen and Dessy	e-nose	[29]
1982	First electronic nose	Persaud and Dodd	e-nose	[27]
1985	Multisensor array for liquid phase analysis	Otto and Thomas	e-tongue	[21]
1988	Definition of an electronic nose	Gardner	e-nose	[27]
1992	Taste sensor	Toko	e-tongue	[22]
1995	Electronic tongue	Vlasov, Legin, D'Amigo, Di Natale	e-tongue	[23]

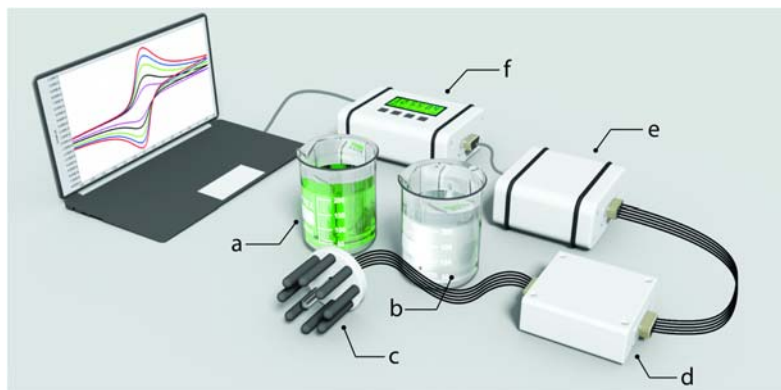


Fig. 7.3. Rendered view of electronic tongue setup: a – sample; b - electrode rinsing solution; c - array of electrodes; d - relay box; e – multiplexer; f – potentiostat.

Electronic tongues can be set up as either desktop or mobile versions. They can also be either static, i.e. when the sensors are submerged in static liquid, or dynamic, with the liquid pumped through the sensor chamber. There is usually also a second chamber used to rinse the sensor array after each analysis. The liquid containing analytes is often thermostated in order to maintain stability of measurement and repeatability. The types of chemical sensors used in electronic tongues do not differ in a significant way from the previously discussed sensors employed in electronic noses. One distinct category of devices are the so-called ‘bioelectronic tongues’, in which enzymes selective for particular compounds are utilized [33]. Arrays of biosensors are characterized by high selectivity due to enzyme-substrate interactions. Their efficiency can be further improved by facilitating the transfer of electrons from enzymes to the electrode (Downey). For that purpose electron mediators are introduced to the liquid. Devices equipped with arrays of biosensors have been used to study, among others, alcoholic beverages [34], fruits [35] and aging process in beer [36]. Besides biosensors, potentiometric and impedimetric sensors are also used in electronic tongues. The former are based on the voltage measurement at null current, which is needed in order to retain the balance of electrochemical processes [37]. They are relatively inexpensive and easy to manufacture, and also can be selective. On the other hand, fluctuations of temperature have a significant impact on their response signal. They are also prone to changes in potential caused by adsorption of components of solution on their active surface [6, 27, 39].



7.5. Sensors Used in Electronic Noses and Electronic Tongues

During measurement process an initial form of energy (an input signal) is transformed into a different form of energy, such as: electric, magnetic, chemical, thermal or radiation energy, which constitutes an output signal [39]. The element using which the measured parameter is read is called 'senor'. Together with certain type of signal converter enables the process of described transformation [28, 40]. Sensor is the most important and primary element in electronic noses and electronic tongues. There are two main critical properties of sensors, which makes them more or less useful in dedicated application. It is sensitivity and selectivity to odorants or taste substances that may be present in the analyzed samples. Sensors can be divided into four groups, i.e. electrochemical (conductometric, potentiometric, amperometric, voltammetric, and impedimetric), piezoelectric, optical sensors and biosensors. In case of electronic noses, instead of array of chemical sensors the proper detection system can be also applied. Some of this systems are equipped with gas chromatograph and/or universal detector, e.g. flame ionization detector, ion mobility spectrometer or mass spectrometer. Sometimes as a substitute of the detector the SAW sensor can be applied [27].

7.5.1. Conductometric Sensors

Conductometric sensors is not very often used in electronic tongues [33]. Their use is primarily in electronic noses. Their principle of operation is based on the changes in conductivity. These changes result from the interactions with the volatile odorants which leads to the changes in the sensor's electrical resistance. Despite the fact there are various types this sensors, the construction and distribution of specific elements in conductometric sensors is, in principle, the same. Conductometric sensors can be divided to types, i.e., Metal Oxide Semiconductor (MOS), Metal-Oxide Semiconductor field-Effect Transistor (MOSFET) and Conductive Polymer (CP) sensor [18, 28].

By far the most commonly used type of sensors are metal oxide semiconductors (MOS). They are popular because of their durability, relatively low price, sensitivity in the range from 5 to 500 ppm, chemical resistance and ease of use [42, 43]. Their active components are usually made of zinc, tin, tungsten or iridium oxides doped with platinum or palladium. When an analyte comes into contact with the active surface the sensor's resistance changes due to a catalytic reaction. The operating



temperature of MOS sensors is in the range of 300 °C-500 °C which shortens the response and recovery time and prevents water vapour from condensing on their surface [15]. Unfortunately, this results in high power consumption, which is why this type of sensors is rarely used in portable devices. Moreover, they are susceptible to poisoning with sulphur compounds which create stable bonds with metal oxides [7, 26].

Metal oxide semiconductor field-effect transistors (MOSFET) are similar to MOS sensors, but operate at lower temperatures (100 °C - 200 °C). They are manufactured by depositing a catalytically sensitive metal in a gaseous phase on a silicon oxide surface. When analytes come into contact with the catalyst a reaction takes place, during which the sensor's physicochemical properties are modified, which in turn leads to a change in electrical potential. The sensors response signal magnitude is proportional to the concentration of analytes in the gaseous mixture. It is possible to manufacture very small MOSFET sensors without sacrificing the repeatability of analysis [44]. Due to the fact that the number of MOSFET varieties available on the market is relatively small they are seldom used in commercial devices.

Sensors based on conductive polymers (CP) belong to the so-called cold conductometric sensors. They are made of semiconducting materials set between two gold-plated electrodes. After the interaction with volatile molecules, reversible changes in conductivity are observed in the sensor. CP sensors are mainly sensitive to polar compounds. However their selectivity and sensitivity can be modified by introducing different functional groups to the polymer structure [17, 45]. Due to such modification, polymeric or glass fiber composites may also display high sensitivity to non-polar substances [46, 47]. The advantages of CP sensors are a low price and fast response, while susceptibility to humidity is the main disadvantage. These sensors have been used in an electronic nose to, inter alia, identify the stages of wine fermentation, monitor decomposition in the Atlantic salmon during its storage at different temperatures and detect the spoiled vacuum-packed beef. Cyranose 320, Aroma Scan A32/50S and Bloodhound BH114 are commercially available electronic noses that employ CP sensors [48].

7.5.2. Potentiometric Sensors

Potentiometric sensors are main elements of electronic tongues. These devices are used to monitor cheese fermentation [49], evaluate the impact of micro-oxygenation and oak chip maceration on the wine



composition, in particular on the presence of phenol compounds [50], monitor changes during beer brewing [51], and identify the botanical origin of honey [52]. This principle of operation is based on the voltage measurement at null current, which is usually needed to retain the balance of electrochemical process. The generated signals are the electromotive forces which depend on the analyte activity. These signals originate in the process of electrochemical reaction at the electrodes or in solution. Potentiometric sensors have a lot of advantages, such as: low cost, commercially easy to produce, possibility to obtain selective sensors, and the highest degree of similarity with the mechanism of molecular recognition. Main disadvantages of this sensors are resulted by the dependence of the measured value on temperature and adsorption of the solution components onto the electrodes [52].

7.5.3. Amperometric and Voltammetric Sensors

This type of sensors is often used in electronic noses. Their use enables to assign the correct class to wheat samples in accordance with the quality classification [53]. They are also applied in electronic tongue systems to assess the conditions under which olive oil is stored [54], identify white wines with regard to the type of grapes and geographical origin [55], and discriminate among various blends of fruit juices [56]. Additionally, similarly to potentiometric sensors, these sensors are used to monitor the aging phase of wine [57], monitor beer fermentation [57], control the freshness of milk stored at room temperature [58], detect chemically adulterated red wine [59], and identify rice wine with regard to its age [60]. The measurements by means of these sensors are based on the electric current reading between the working and reference electrode in an electrochemical cell, as a function of analyte concentration. When an analyte reacts electrochemically, i.e. it undergoes oxidation or reduction, the electric current is generated at the working electrode. The reaction usually takes place at a constant potential as controlled with a potentiostat [28, 40]. In general, the working electrodes are made of gold, iridium, palladium, platinum and rhodium [61]. The main disadvantage of such sensors is the lack of selectivity.

7.5.4. Impedimetric Sensors

Impedimetric sensors are usually employed in an electronic tongues, however it does not happen often. This type of sensor was used to



discriminate brands of red wines [60]. Their principles of operation is based on measuring the impedance at one constant frequency or for a frequency spectrum by means of impedance spectroscopy [63, 64].

7.5.5. Piezoelectric Sensors

The working principle of piezoelectric sensors is based on a piezoelectric phenomenon. This group can be divided into surface acoustic wave (SAW) sensors and bulk acoustic wave (BAW) sensors which also include quartz microbalance sensors (QCM). The sensors mass changes when analytes adsorb on its surface, which in turn leads to change of frequency at which the measuring element resonates [14]. This type of sensors is commonly used for measuring temperature, pressure, force or acceleration [17, 65]. The main difference between BAW and SAW sensors is, that in the former the generated wave travels through its entire volume, while in the latter only along its surface [17]. The main advantages of piezoelectric sensors are high sensitivity, real-time measurements, small size, durability, low cost. Piezoelectric sensors are commonly used in electronic noses to determine the optimal time for harvesting apples [65] and evaluate the quality of tomatoes [66]. Their wider use in electronic tongues is just a question of time.

Bulk acoustic wave sensors are comprised of a single quartz crystal and two gold-plated electrodes [15]. The crystal is coated with a selective layer which absorbs measured chemical substances from the gaseous mixture. When this happens, the resonator's weight increases and its resonating frequency, which ordinarily oscillates between 5 to 30 MHz changes. This gives rise to a three-dimensional acoustic wave which passes along the entire bulk of the crystal. Purging the crystal with an inert gas causes desorption of analytes and a return to default frequency [67]. The use of QCM sensors has numerous advantages, namely high sensitivity (ppb level), linearity of response at a large range of concentrations and insusceptibility to fluctuations of temperature and relative humidity.

Surface acoustic wave sensors have a higher resonating frequency of app. 100 MHz – 1 GHz. In this type of sensors a piezoelectric material with input and output transducers situated at its surface is used. A selective membrane is placed between the transducers. The membrane is usually made of porous polymers, lipids, self-assembled monolayers or Langmuir-Blodgett films. A two-dimensional acoustic wave travelling along the sensor's surface is generated when alternating current is



directed through the input. The piezoelectric element is made of zinc oxide, lithium niobate or quartz. SAW sensors are less sensitive than QCM, since they react to concentrations at ppm level, but their response time is relatively short [15].

7.5.6. Optical Sensors

The use of optical sensors makes possible to detect substances which are not electrochemically active and therefore cannot be detected by an electrochemical sensor. Optical sensors have some disadvantages which significantly limit their application, for example, sensor durability and the output signal distortion [67, 69]. These sensors have been employed in electronic noses to discriminate commercial drinks [68], while in electronic tongues to evaluate the quality of beer brands [69] and discriminate wines with regard to the wine age and grape variety [59]. Optical sensors are devices mostly based on the interaction of electromagnetic radiation with matter. These sensors contain various dyes which react with analytes. The sensor sensitivity depends on the type of dyes used, a plethora of dyes can be employed in electronic senses [46, 70]. Optical sensors can be based on different phenomena such as, fluorescence, reflection and absorbance. These sensors consist of an indicator, detector and the light source, the latter set at a specific wavelength in order to maximize selectivity [17, 18]. The sensors are placed on special, mostly polymeric-type membranes. Due to the interaction with an analyte, the properties of an indicator change, which influences the membrane absorbance or fluorescence. The changes are monitored via a detector, which converts the signal from optical into electric form. There is a big variety of optical sensors therefore they are characterized by low cost, simple procedure and high selectivity.

7.5.7. Biosensors

Biosensors are mainly used in electronic tongues. These sensors have been used to monitor changes occurring during the aging process in alcohols [71]. A biosensor consists of a biological measuring element, which is located close to the transducer in order to achieve high sensitivity to the target analytes (Fig. 7.4).

Biosensor-based electronic tongue is often called a bioelectronic tongue. Until now, different principles of operation of bioelectronic tongues have been proposed, including voltammetric, amperometric and



potentiometric principle. One of the first voltammetric biosensor comprised three biocomposite electrodes that contained glucose oxidase and various metals such as, platinum, palladium and silver [71]. The next developed biosensor was based on the combination of potentiometric sensor and the enzyme urease, which had been covalently bound to carboxylated poly(vinyl chloride) [42, 63]. Another proposed solution was to combine the amperometric sensor, based on carbon and platinum working electrode and Ag/AgCl reference electrode, and the enzymes cholinesterases, tyrosinases, peroxidases and cellobiose dehydrogenases [62]. Biosensor arrays display high selectivity due to enzyme-substrate interactions. Moreover, the biosensor efficiency can be improved by the introduction of electron mediators, which facilitate the transfer of electrons from the enzyme to the electrode [72].

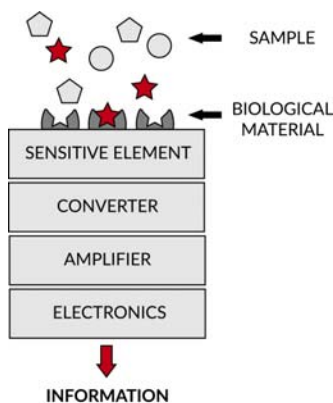


Fig. 7.4. Construction of biosensor.

7.6. Data Analysis Methods

The dataset obtained with the use of electronic nose or electronic tongue contains the response signals of each sensor and usually is very complex. Analysis of this type of data is considerably more difficult, than in the case of a device equipped with only one sensor. For that reason the first step of data processing is usually meant to decrease the dimensionality of the dataset. Doing this whilst retaining as much significant information as possible is one of key challenges in statistical data processing, as the results of data analysis should lead to reliable and repeatable results. Chemometric methods used for data processing utilize pattern recognition. Information contained in the sensor's response



signal is compared with reference data. The basic steps of data analysis are as follows [17, 74]:

- Pre-processing;
- Selection of variables;
- Classification;
- Decision making.

Preliminary analysis (pre-processing) is used to smoothen the signal, average sensor responses, and to filter the background noise [74]. Moreover, at this stage operations like data normalization and centering are also performed. Next step is to reduce the dataset's dimensionality and to employ one or more data analysis techniques. These techniques can be classified into quantitative methods and pattern analysis [76, 77] or, based on the model's learning mechanism into supervised and unsupervised methods. In supervised techniques, in order to properly 'teach' the model, it is fed input values together with expected outputs. In this way a reference signal is defined. In unsupervised methods the model is built without pre-defined outputs.

One of the easiest data analysis methods is to plot raw data as histograms or radar plots. This method works well when there are significant differences between groups of samples [74, 78, 79]. When it is difficult to classify samples outright, methods like multifactorial analysis or neurocomputers are employed [80–82]. In this chapter, the briefly characterization of the most commonly used statistical data analysis methods in combination with electronic noses and electronic tongues are presented.

7.6.1. Artificial Neural Networks (ANN)

Models utilizing artificial neural networks can work both as supervised and unsupervised pattern recognition techniques. Artificial networks are comprised of interconnected computation clusters, so-called 'nods'. In each node a simple mathematical operation that modifies the input is performed. This solution mimics the processes taking place in human neural cells. Nods are usually situated in layers. The task of the first strata is to relay the information to other nodes. Subsequent nodes modify the signal and their output is the input of other clusters. In this way, neural network 'learns' how to react to a given impulse, e.g. a mixture of odour compounds [82]. At this stage weights are determined, that is by what factor the signal will be multiplied in a given node. Nodes situated in the



last layer are called output neurons. A model set up in this way can be used to predict output values, classification or for pattern recognition [83]. The most commonly used Chemometric methods based on ANN are radial basis function (RBF), counter propagation – artificial neural network (CP – ANN), generalized regression neural network (GRNN), time delay neural networks (TDNN), probabilistic neural networks (PNN) and self-organizing maps (SOM) [84].

7.6.2. Cluster Analysis (CA)

In this method inputs are classified based on the distances between individual data points [85]. The result of cluster analysis is often plotted in the form of dendrograms [86]. Each strand is assigned to gradually larger agglomerates based on the similarity of data (distance between particular strands). In the end each object forms a single cluster [85]. Clusters can be grouped not only based on distance to the nearest neighbour, but also based on the distance from the furthestmost neighbour or group average [86]. Each object in the dataset can be assigned to only one class. In practice, CA can be used to determine, whether or not there are distinct sub-classes in a given group [87].

7.6.3. k-Nearest Neighbour (k-NN)

K-nearest neighbour algorithm classifies data points based on distances between them in a multidimensional space. The points that are closest to each other create a separate group. Input data is compared with regard to Euclidean distances and therefore k objects that are most alike are selected. The 'k' parameter is chosen in such a way, as to maximize class separation [88].

7.6.4. Discriminant Function Analysis (DFA)

In this method discriminant functions are defined in such a way as to maximize the variance between various classes, and at the same time minimize the distances within the classes themselves [89]. This is achieved by using discriminant prediction equation, which allows to discard the variables that have the least impact on classification. In practice, first the F-test is performed in order to determine whether the discriminant model is entirely significant, and subsequently when the F-test has shown significance [90].



7.6.5. Partial Least Squares (PLS)

Partial least squares method is a modification of principal component analysis. It is a good example of a multiple regression method. The algorithm tries to fit a linear combination of predictors. These predictors, just like principal components, are orthogonal to each other. New variables have to explain the input data and be correlated with output values. PLS is employed when a single response signal can be explained by a large number of variables. It can be used to develop discriminant analysis models (PLS-DA) [91].

7.6.6. Linear Discriminant Analysis (LDA)

A probabilistic classification technique, LDA is based on rotating an orthogonal data space in such a way, as to maximize the variance between classes, and minimize the variance within those classes [82], [87]. Classes should have a normal distribution, there should be a linear separation of groups and variance-covariance matrices, and the number of objects should be at least three times larger than the number of variables [92, 93]. At the model training stage outputs of classes are established, add then input data is assigned to these classes [87].

7.6.7. Soft Independent Modelling of Class Analogies (SIMCA)

In SIMCA a separate model is created for each class. To this end e.g. principal components can be used [92, 94]. Then, based on a pre-defined similarity, a confidence envelope is created which allows to determine whether an object belongs to a particular group or not. The number of principal components used to create a model varies with each class.

7.6.8. Support Vector Machines (SVMs)

Support vector machines is an increasingly popular method of data processing in electronic noses. It was first proposed by Vapnik in 1992 [95], which makes it one of most recent supervised learning techniques. In it, points in a multi-dimensional space are separated by a hyperplane to form distinct classes. How successful this separation is depends on hyperplane's confidence margin, defined as the distance between the hyperplane and object closest to it (so-called support vectors) [96, 97]. Variants of this method are also used, such as support vector regression or least squares support vector regression (LSSVR) [98].



7.6.9. Analysis of Variance (ANOVA)

This method is used to compare more than two groups, classification of which is based on a single variable (one-way ANOVA). In this parametric tool the variance of dependent variables is compared within groups which were created based on independent variables. In order to employ the analysis of variance it is necessary to know the dependent variables that are characterized by the normal distribution and by being measurable at least on an interval scale. Moreover, it is important for the groups of variables to be comprised of the same number of objects [99].

7.7. Applications

To date, a great number of studies devoted to various applications of electronic noses and electronic tongues are reported. This artificial senses were used in many different areas of everyday life, e.g. in chemical industry including explosive materials, petrochemical, cosmetic, pharmaceutical, paper, packaging industries, as well as liquid-gas distribution and bootling plants, criminological and medical tests, agriculture, transportation, manufacturing and military purposes. In respect of many years of using this technology the general conclusions can be deduced. Firstly, rather more applications are devoted to electronic noses than electronic tongues and secondly, the main area of interest of artificial senses during many years and now is still food industry. Second main application is environmental monitoring, which was the first use of electronic sense, namely electronic nose to analyze the mixtures of volatile air contaminants that had already been detected by olfaction [72]. Therefore, in present subchapter this two above-mentioned and key applications will be discussed, despite of the fact that electronic tongue utilization in second application is minor.

7.7.1. Food Investigations

In the case of food process monitoring, the electronic nose and tongue have been used to monitor fermentation in milk and cheese [50, 101]. The application of artificial senses assures the product quality at the very start of the food production line. The monitoring of food freshness and product quality during storage is the foremost application of the electronic nose and electronic tongue [32, 59, 81, 102–109]. This allows



the exclusion of spoiled products from the market as well as the determination of appropriate shelf times and storage conditions for milk and cheese in order to avoid financial losses by the dairy industry. Chemical analyses of meat products are mainly performed via an electronic nose, the use of electronic tongue in this case is less suitable because it requires a more complex preparation of samples [31], medical condition of meat [102, 103], and the influence of storage conditions on meat quality in order to determine expiration date [111]. The electronic tongue was applied to analyze ground meat in order to predict the level of chlorides, nitrates and nitrites [110]. Actually, it is the electronic nose which has found a broader application in the field of meat product analysis. This device is used to, among others, monitor the curing process in Iberian ham in order to detect spoilage Fish and shellfish [111]. The electronic senses are employed in the fish processing industry to mainly evaluate the freshness of fish and shellfish. The electronic nose has been used to perform such analysis in sardines [112, 113], shrimp and cod roe [59, 72], and the Atlantic salmon [114], while the electronic tongue, on samples of bream [115]. The duration and conditions of storage have big influence on the freshness of fish and shellfish. In order to protect consumers from the purchase of old fish, studies on the relationship between the duration and conditions of storage, and product freshness were conducted by means of an electronic nose on cod fillets [116], fresh and frozen Atlantic salmon, tilapia [116], the Argentine hake [117], and oysters [118]. An electronic nose is also used to analyze fruits and vegetables. The application of this device is related to the monitoring of food processing, and the evaluation of freshness, shelf life and authenticity of food. On the other hand, the electronic tongue has been mostly used to classify cultivars (e.g. discrimination between onion and shallot) [119], tomatoes based on various parameters [120], apples [121] and apricots, the latter being also discriminated based on the storage duration [121]. The following fruits and vegetables have been investigated so far: bananas, apples, pears, oranges, strawberries, broccoli, potatoes and carrots. The electronic nose was used to monitor dehydration in tomatoes [122] and grapes [123] in order to determine the optimal storage time. Olive oil is very frequently analyzed because the attempts to adulterate more expensive olive oil with its cheaper alternative are very common [123]. Therefore the electronic tongue and nose were both used to discriminate among olive oils based on oil geographic origin [124], and type and quality [55, 125]. In the case of olive oil, the conducted studies were also aimed at determining rancidity [126] and the relationship between the storage time and oil quality [54]. In the case of grains and grain products, quality evaluation and



authentication are important (wheat [53, 29], rice [127], barley and oats [128, 129], and corn [130]) however the critical issue for the consumers is the evaluation of possible health risks of such products by means of electronic senses. The possible health risks are related to grain diseases, which often go unnoticed due to the fact that they are not visible to the naked eye. Mycotoxin contamination of grain, which also includes contamination with aflatoxins, and various diseases caused by fungi and bacteria are such risks. Coffees, teas and herbal infusions are mainly analyzed in order to distinguish among specific types, quality levels and brands. This is due to the fact that these products are highly variable. The products of low and high quality are frequently mixed together to lower the overall production costs, and then sold as top quality merchandise. Coffees were discriminated based on the quality [131], brand [119, 132, 133] and the ripening period [134], while teas were evaluated in relation to quality [135], brand [136], geographical origin [136], and the content of flavoring substances such as, coumarin [137], theaflavin [138]. Moreover, the electronic nose was used to monitor the fermentation process in black tea, and to determine the optimal time for producing tea with the best flavor [138]. Until now, the studies aimed at identifying brands and quality of beverages were conducted in e.g. cola type drinks [138], other commercial beverages [139], mineral water [140], and fruit juices and fruit juice-based drinks [141, 142]. On the other hand, alcoholic beverages are among the products which have been most frequently analyzed by means of electronic senses. Process monitoring in alcohol production requires fast analytical tools which can detect substandard products, discriminate among products, and authenticate products in real-time. Wine and beer is mostly subjected to such type of monitoring because both beverages undergo fermentation that results in a release of specific compounds influencing the taste and aroma of the final product. The artificial senses monitored the processes of fermentation [142], brewing [51] and aging [71] in beer. In the case of wine, these devices were employed to control the aging process, determine the influence of wooden barrels on aging and maceration [51, 58], and monitor grape fermentation [143]. Authentication of alcohols is not only aimed at detecting adulterated products with substandard characteristics, but also at identifying falsified products that can be potentially harmful to the consumer's health. Until now, the electronic nose was used to discriminate among vodkas [144], spirits [145], whiskeys [146], wines [146], tequilas [146], beers [146] and sorghum-based drinks [147]. The examples of electronic nose and electronic tongue applications in food analysis are shown in Table 7.2.



Table 7.2. Application of electronic noses and electronic tongues in food investigations.

Application	Sample	Object of investigation	Method of data analysis	Ref.
Food process monitoring; E-NOSE	Milk and cheeses	Monitoring of the smell intensity during fermentation	PCA	[148]
	Cencara tomatoes	Monitoring of the dehydration process	PCA	[122]
	Black tea	Determination of the optimal duration of fermentation	TDNN, SOM,	[149]
Evaluation of food freshness; E-NOSE	Cod fillets	Discrimination of samples based on different storage times	PLS	[116]
	Smoked Atlantic salmon (fresh and frozen)	Classification of spoilage in samples at different temperatures	PLS, PLSR	[114] [150]
	Meat	Freshness evaluation in meat in relation to storage time and storage conditions	PCA, LDA, BPNN, CDA	[150]
	Olive oil	Detection of rancidification	PCA	[151]
Testing the shelf life of food; E-NOSE	Pink Lady and Jonagold apples	Discrimination of varieties ripeness level, shelf life and storage conditions	PCA, ANN, PLS,	[152] [153] [154]
	Milk	Determining the influence of storage time on milk	PCA,	[105] [155]
	Meat	Determination of shelf life	LDA	[101]
Authentication of food; E-NOSE	Tequila, whiskey, vodka	Discrimination of four types of beverages	PCA, DFA	[146]
	Italian wines	Detection of the falsification	PCA, BP/ANN	[156]
	Green tea	Discrimination of the quality classes in Longjing tea	LDA, PCA,	[157]
			ANOVA, ANN	[135]
		Identification of coumarin-enriched green tea	PCA, CA	[137]
Discrimination of the green tea brands	PCA, ANN	[137]		
Other applications; E-NOSE	Olive oil	Discrimination of quality classes based on the qualitative and quantitative information	SOM,	[137]
			SIMCA, PLS	[158]
			PCA	[159]
	Cabernet red wine	Monitoring of changes in wine aroma after bottle opening	PCA, SOM	[160]
Oranges, apples and peaches	Post-harvest quality evaluation	PCA, PLS, ANN	[161] [162]	
Food process monitoring; E-TONGUE	Bacterial cultures used in the cheese production	Monitoring of the fermentation process	PLS	[49]



Application	Sample	Object of investigation	Method of data analysis	Ref.
	Red wines	Monitoring of wine aging	PCA, SIMCA	[163]
		Evaluating the influence of micro-oxygenation and oak chip maceration on wine composition.	PCA, PLS, ANOVA	[50] [164]
	Beer	Variability monitoring of the brewing process	PCA, PLS	[51]
		Monitoring of changes during the aging process in beer	PCA, LDA, RBF, PNN, BP-NN	[51]
		Monitoring of the fermentation process	PLS	[69]
Evaluation of food quality and freshness; E-TONGUE	Fillets of farmed gilthead seabream <i>Sparus aurata</i>	Discrimination of storage time. Predicting the parameters of spoilage	PCA, ANN, PLS MLR	[165]
	Tench (<i>Tinca tinca</i>)	Discrimination of storage time	PCA	[166]
	Apricots	Discrimination based on post-harvest storage time and apricot varieties	CDA, PLS	[166]
	Non-alcoholic beverages	Quality evaluation of high-fructose corn syrup	SIMCA	[51]
	Rice	Quality evaluation of milling	PCA	[167]
	Milk	Monitoring the freshness of milk stored at room temperature	PCA, ANN, CA, PLSR, LS-SVM	[58] [168]
Stability testing of food; E-TONGUE	Extra virgin olive oil	Evaluation of different storage conditions	LDA	[54]
Authentication; E-TONGUE	Honey	Discrimination based on botanical origin	PCA, PLS, ANN, LDA	[52] [169]
		Discrimination based on botanical and geographical origin	PCA, CA, CCA, ANN	[170] [171]
	Milk	Discrimination of milk samples pasteurized in different ways	PCA	[171]
	Alcoholic beverages	Fast quality evaluation of alcoholic drinks and identification of brands	PLS, LDA, PCA	[172]
		Determination of ethanol content in alcohols from different sources	PCA, SIMCA, PLS, PCR	[173]
		Discrimination between the cheap and expensive whiskies	PCA	[174]
Other applications; E-TONGUE	Extra virgin olive oil	Discrimination of samples based on phenolic compounds content	PCA, PLS, PLS-DA	[175]
	Black tea	Determination of theaflavin level	PCA, ANN	[138]
	Ground meat	Prediction of the level of chlorides, nitrates and nitrites	PLS	[138]

7.7.2. Environmental Monitoring

Electronic senses, namely electronic nose and electronic tongue, can be successfully used for monitoring environmental pollution. There is a need to develop devices capable of evaluating the state of the environment rapidly or even in real time, without supervision and at a relatively low cost. Electronic noses and tongues show promise in this regard and find application in monitoring the quality of water and of atmospheric air [176, 177]. The analysis of atmospheric air can be performed in several ways. One can, for example, measure the concentration of several pre-defined substances or analyse the air holistically. Both these tasks can be performed using sensor systems. One of the first investigations in this area was performed in the first half of 1990' [178]. In it, a device equipped with CP sensors was used to analyse an aqueous solution of ethanol, diacetyl and dimethyl sulphide. Electronic nose was also used to measure the concentration of nitric oxide, methane and carbon monoxide at 500-2000 ppm concentrations [179]. It is important to note, that when using e-noses to determine particular substances there can occur interferences caused by the presence of other chemical compounds. A research has shown, that when determining hydrogen sulphide and nitrogen dioxide in a mixture containing carbon dioxide and water vapour the presence of humidity and CO₂ had a significant impact on the sensor's response signal, but it was possible to properly identify the components of the gaseous mixture using discriminant factor analysis [180]. It is possible to use the electronic nose to determine certain VOCs at a very low concentration level (ppb level) [181, 182], even below the threshold limit value (TLV). That was the case with benzene, methanol, ethanol, toluene and acetone determined below TLV using a device equipped with MOS sensors [183]. Sensor drift poses a significant problem, leading to high measurement uncertainties. In most recent applications electronic noses are being mounted on mobile robots [184], but because of insufficiently advanced models of gas distribution this technology is yet to find real-life applications. Another important application of electronic noses is odour classification and odour intensity evaluation. Currently, the golden standard in determination of odour nuisance is dynamic olfactometry [185]. Using this technique it is not possible to determine the nature of the odorant. Moreover, it is not sufficiently sensitive and cannot be used to constantly monitor odour nuisance [177]. A solution can be to use sensor systems. In one of the first investigations in this area two electronic noses: Odourmapper and Aromascan PLC were used for odour quantification [186]. When investigating odour intensity, the results



obtained using e-noses are similar in nature to those obtained using dynamic olfactometry. In one study that compared the two techniques when applied to poultry livestock nuisance monitoring has shown, in the range from 250 to 4500 ou/m^3 a linear correlation with R factor of 0.89 [187]. Classification of odorous air is also an important issue. Often an unpleasant odour, even at low intensity, is less acceptable than a pleasant one at high intensity. In 2007 the use of electronic noses for classification of odours in the vicinity of a composting plant was evaluated [188]. The accuracy index of classification was 72 %. Electronic olfaction can also be used for evaluation of water quality, e.g. to determine the biological oxygen demand (BOD) [189, 190]. Electronic noses were also used to determine pesticides [191], sulphides and nitrates [192] or to detect the presence in water of microorganisms responsible for emission of odours [193]. Other applications of e-noses in environmental protection include the control of processes taking place during composting or sewage treatment and monitoring of the operation of devices used for purifying water and gasses from undesirable substances, including odours [177]. The examples of electronic nose applications in environmental monitoring are presented in Table 7.3.

Due to their limitations electronic tongues are not used as often as electronic noses in environment monitoring. The main area of their application in this area is detection of water pollution, both potable and processed at sewage treatment plants. Electronic tongues can be used e.g. to monitor semi-volatile compounds like herbicides [176] or determine the contents of inorganic anions and transition-metal cations in model ground, mine and sea water.

7.8. Summary

Over last 30 years the evolution of sensor technologies gives a great opportunity to apply a newest technical solutions into the systems where sensor arrays are utilized. Electronic noses and electronic tongues in the assumptions are devices dedicated to measure aroma and taste in a way parallel to the human biological senses. This gives a great opportunity to apply them where human being is not able to make proper measurements with required repeatability, objectivity and sensitivity, and in the places where toxic conditions may exists. Due to the fact that these devices are electronic ones, it gives a huge potential for applications in which *on-line* or data transmission systems is considered.

Table 7.3. Examples of the application of electronic noses in analysis of air pollution.

Type of sample	Analytes	Data analysis method	Ref.
Air samples from composting plants, printing houses, sewage treatment plants, recycling plants, settlers	Water vapor, flammable gases, toxic gases, solvents	PCA, DA	[194]
Internal air samples	CO, NO ₂ , VOC (benzene, toluene, m-xylene)	SOM	[195]
Samples of air from the sewage treatment plant	VOC	PCA	[196]
Air samples from sewage treatment plants	VOC	SOM	[197]
Indoor air samples from duck farms	H ₂ S, NO ₂ , SO ₂	DFA	[198]
Internal air samples	Ammonia, VOCs	ANN	[199]
Internal air samples	CO, NO ₂ , ammonia, VOC (benzene, toluene, formaldehyde)	HSVM, SVM, FDA, MLP	[200]
Samples of indoor air from cars	CO, NO ₂ , VOC (benzene, toluene, formaldehyde)	BPNN	[201]
External air samples of from Alexandria	CO, CO ₂	ANOVA	[177]

This fits into contemporary canvas of national and international smart specialization strategy, especially when the use of telemedicine is expected. Moreover, these systems have even more advantages of which the possibility of their miniaturization and execution of rapid analysis is the key one. Therefore, despite of the fact that there are already many applications of electronic noses and electronic tongues, even more can be invented in the known areas and new ones. For example, the biosensors are more commonly used in electronic tongues than in electronic noses. Continuous evolution of sensor technology might, in the near future, lead to balance of this proportions. Furthermore, electronic nose and electronic tongue technology is on the verge of renaissance. It is understandable that the fastest progress in sensor development was years ago, nevertheless this technology is before the greatest development. The situation is analogical to one of the 50's. That time only a best specialist had a contact with computers. Nowadays, computer is well known device and available at every step. Perhaps the appropriate adaptation of artificial senses to the needs of the average user



(not R & D person) will launch a new chapter in the field of electronic nose and electronic tongue technology.

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