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An electronic nose system to diagnose illness

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Abstract

Recently, medical diagnostics has emerged to be a promising application area for electronic noses (e-nose). In this paper, we review work carried out at Warwick University on the use of an e-nose to diagnose illness. Specifically, we have applied an e-nose to the identification of pathogens from cultures and diagnosing illness from breath samples. These initial results suggest that an e-nose will be able to assist in the diagnosis of diseases in the near future. © 2000 Elsevier Science B.V. All rights reserved.

Keywords: Electronic nose; Medical diagnostics; Neural network

1. Introduction

Electronic noses (e-noses) have been used to analyse the complex odorous headspace of food and drink — mainly employing metal oxide or conducting polymer resistive gas sensors as shown in Table 1. The table summarises some of the reported applications of e-noses (research and commercial) pertaining to the food industry which are listed from seafood, such as oysters and squid, through to cheeses, such as cheddar. The applications usually involve the grading of the odor in terms of the type of foodstuff or its quality (e.g., freshness). Thus an e-nose may help assure the quality of both processed and raw foodstuffs.

Monitoring the spoilage of foodstuffs is closely related to the monitoring of the growth of bacteria in a certain medium. In other words, an e-nose may be able to recognise characteristic smells from diseases and bacteria cells because cell metabolism is the biological oxidation of organic compounds, such as glucose ($C_6H_{12}O_6$), to yield ATP and secondary metabolites as shown in Fig. 1. There is now great interest in the clinical application of an e-nose; that is in the ability of an e-nose to diagnose illness. It is well known that certain diseases are associated with characteristic smells, for example, diabetes produces the sweet smell of acetone on the breath and stomach ailments are often associated with halitosis. Other diseases, like cancers associated with the lungs, liver and intestine can also produce characteristic odors. Some reported clinical applications of e-noses are listed in Table 2.

We now describe work carried out at Warwick University on the use of an e-nose for three medical applications: (i) the identification of pathogens that cause infectious disease of the upper respiratory tract and ears; (ii) the classification of cyanobacteria, *Microcystis aeruginosa*, in water that may produce toxins that are poisonous to cattle and people; (iii) diagnosing the presence of sub-clinical or clinical ketosis from the breath of dairy cows. We believe that these three examples demonstrate that there is a role for the future employment of e-nose instrument within the field of medical diagnostics.

2. Experimental

Measurement systems were designed for each of the three applications. Fig. 2 shows the design of a fully automated e-nose system to sample the headspace of the pathogen and record the data. Air is passed over the pathogen sample and passed into a specially adapted e-nose. The e-nose is based on a Fox 2,000 (Alpha MOS SA) and comprises odor sensor arrays. At this stage, we used only six commercial MOS gas sensors (Alpha MOS) in the e-nose.

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Table 1
Some reported applications of e-noses in the food industry

Food	Test	Sensors/type	References
Seafood (oyster, sardine, squid)	freshness	1/MOS[a]	[1]
Fish (cod, haddock)	freshness	4/MOS	[2]
Fish	freshness	1/MOS[a]	[3]
Fish (trout)	freshness	8/EC	[4]
Grains	classification	15/mixed	[5]
Ground pork/beef	discriminate and effect of ageing	15/mixed	[6]
Boar	taints in meat	14/MOS	[7]
Sausage meats	discriminate	6/MOS	[8]
Food flavours (orange, apple, strawberry, grape, peach)	flavour identification	8/BAW	[9]
Wheat	grade quality	4 × 4/EC	[10]
Wheat and cheese	discriminate and ageing	20/CP	[11]
Tomatoes	effect of irradiation and stress	7/mixed	[12]
Cheese	maturity of cheddar	20/CP	[13]
Cheese	discriminate	8/CP	[14]

[a]Strictly not an electronic nose (as defined here) but an odour monitor. Key: MOS = metal oxide semiconductor, EC = electrochemical, BAW = bulk acoustic wave, CP = conducting polymer.

2.1. Identifying respiratory infections

First, we applied an e-nose to identify bacteria that cause disease of ears, nose and throat. Diseases of the upper respiratory system are often associated with microorganisms, such as *Staphylococcus (S.) aureus*, *Legionalla pneumphilia*, and *Escherichia (E.) coli*. The current clinical procedure involves the taking of a sample by swabbing the infected area with a sterile cotton swab, growing the bacteria in a nutrient medium (e.g., blood agar), followed by assaying/staining for classification. The whole process can take many days by which time the disease may have progressed unchecked or antibiotics may be poorly targetted. Thus, there is a need for a more rapid diagnostic technique, perhaps using an e-nose, to detect both the type and growth phase of a bacterial infection. Our instrument has been used to monitor not only a number of different bacteria but also the growth phase of these microorganisms. Clearly, it is clinically advantageous to detect the presence of pathogenic bacteria in their earliest stage of growth (i.e., the lag phase) rather than their latter stages, which may not be reached for several days. The instrument was used to measure the odorous headspace of different bacteria as they grew in a nutrient medium. Hundreds of response vectors have been recorded for the analysis of bacteria, for full details see Ref. [16].

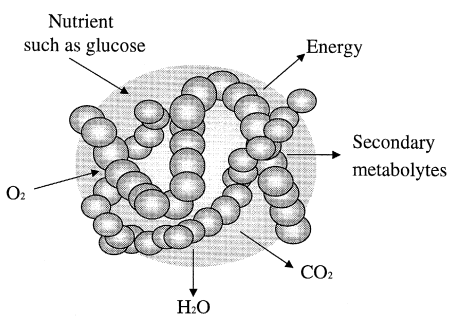

Fig. 1. Schematic diagram of the bacteria cell metabolism.

Fig. 3 shows a linear discrimination plot of the response of the sensor array to four different bacteria. There is clearly some overlap of the response vectors, corresponding to the different bacteria samples, when displayed in linear discriminant space. However, a suitable choice of non-linear pre-processing algorithm and artificial neural network can greatly improve the discrimination.

Table 3 shows the confusion matrix for the prediction of 180 unknown samples (over three different growth phases) using a back-propagation neural network. In this case, 100% of the *S. aureus* samples and 92% of the *E. coli* samples were correctly identified [20]. This is very encouraging because it shows that all of the samples collected during the initial, lag phase of *S. aureus* were correctly predicted — including the first one of these samples, which represented an incubation time of only 10 min or so. A back-propagation neural network was also constructed in order to predict the growth phase of the bacteria. In this case the best success rate was obtained using the absolute final output sensor model and autoscaling (af/a), on average, 80.7% as shown in Table 4. The lower value was due to the uncertainty in the selection of the boundaries between lag/log and log/static growth phases. These were determined by a separate experiment using an optical technique to count the viable cells in the cultures at each time station.

2.2. Identifying water biohazard

Second, we have constructed the measurement system (Fig. 4) for the testing of the cyanobacteria (usually called blue-green algae) which grow in lakes and reservoirs. Cyanobacteria can cause serious nuisance due to unpleasant odors and, in the case of reservoirs, taste. And also, cyanobacteria may produce toxins that are poisonous to

Table 2
Some reported applications of e-noses in medical diagnostics

Pathogens	Application	Sensors	Group	References
6 micros	ENT infections	6 MOS	Warwick	[15]
4 micros	ENT infections	6 MOS	Warwick	[16]
13 micros	various	16 CP	Leeds	[17]
Dietary problem	ketosis in cows	6 MOS	Warwick	[18]
Bacterial vaginosis	vaginal infection	32 CP	UMIST	[19]
E. coli, *S. aureus*	thoracic infections	6 MOS	Warwick	[20]
E. coli, Human factor VII	batch process	MOSFET	Linkoping	[21]

cattle, wildfowl, fish, and people. Thus appropriate methods to detect and quantify these toxins in natural waters is very important. Here we report on the use of an e-nose based on a 6 MOS sensor array to analyse cyanobacteria cultures grown in water with the intent to provide a simple tool to test the water quality as the counterpart of analytical instruments, such as liquid chromatography or optical microscopy. A series of experiments were carried out to analyse the nature of two closely related cyanobacteria, *M. aeruginosa* PCC 7806 that produce a toxin and PCC 7941 that do not. Fig. 5 shows the PCA plot of the second and third principal components of cyanobacteria samples, which indicates two distinct groups of cyanobacteria.

2.3. Diagnosing illness by breath analysis

Finally, we applied an e-nose system to diagnose illness in dairy cows. The dairy industry in the UK is responsible for milk production and the cost of feed for dairy cattle is about one billion pounds per year. Ketosis is a condition in

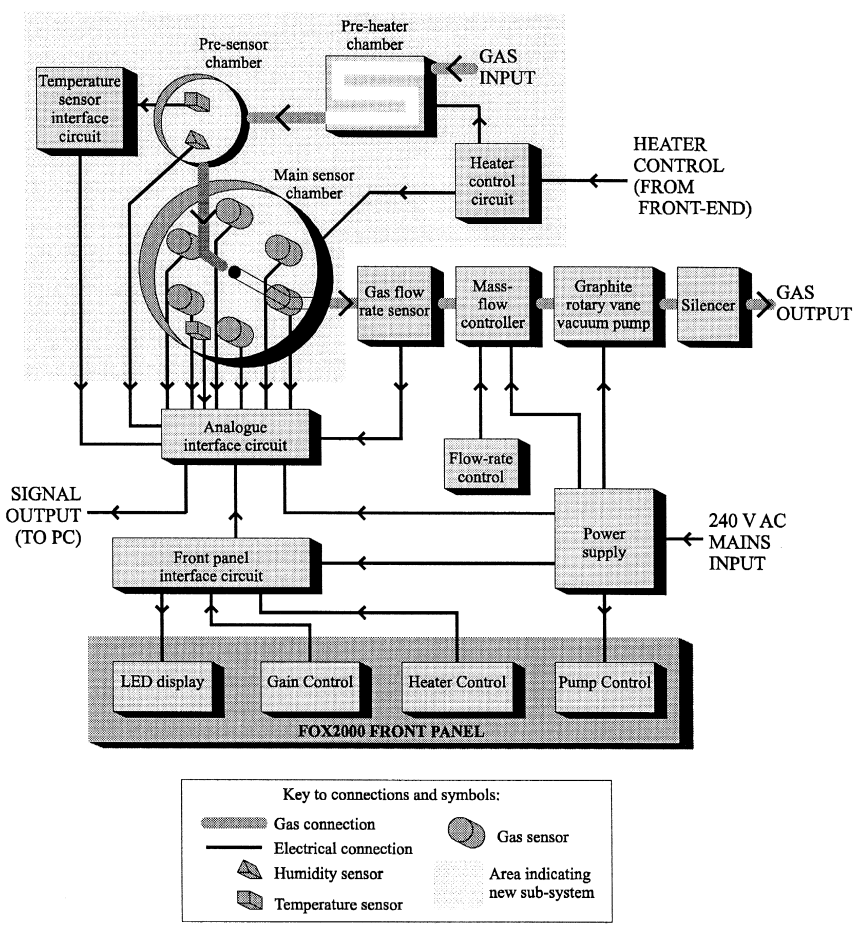

Fig. 2. A modified e-nose system to sample the headspace of pathogen and record the data.

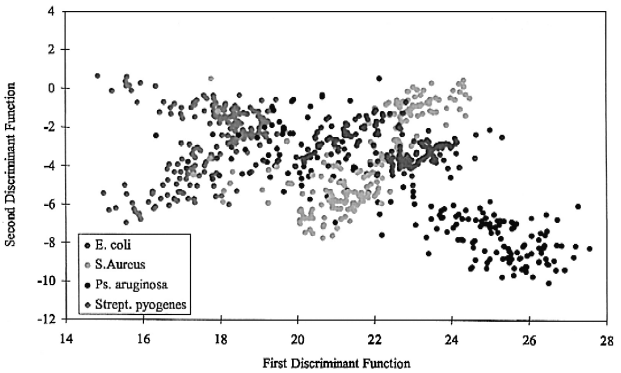

Fig. 3. Recorded responses of MOS-based e-nose to four bacteria samples.

cows associated with the inefficient use of feed and not only causes economic loss but also threatens the health of the cow. Clinical ketosis in cows is detected by the sudden loss of appetite and/or the appearance of the smell of acetone on their breath. Sub-clinical ketosis is normally detected through the analysis of blood or milk samples taken from a cow. However, this is a laborious, costly and intrusive method of diagnosing the health of the cow. Instead, an instrument has been developed in collaboration with Silsoe Research Institute and the University of Southampton to diagnose ketosis in cows [18]. A nasal breath-sampling device was used to collect the breath from cows and store it in a odorless bag. The contents of the bag are then either pumped into an e-nose containing six metal oxide gas sensors or an infrared gas analyser. At the same time, blood samples were taken from the cows by a vet in order to determine the level of 3-hydroxybutanoate (β-HB) concentration that is a good indicator of the ketotic state of the cow.

Table 5 shows the confusion matrix for the prediction of state of health of the cows. The true health of the cows was classified according to the results from the level of β-HB in the blood serum from this matrix. The overall classification rate was found to be 80 to 90% from the e-nose analysis of the breath samples taken. Again, this result is very encouraging and may be explained by the e-nose detecting the presence of elevated levels of propanone in the breath of ketotic cows which were shown to exist by IR results. We therefore suggest that an e-nose can provide a rapid and unobtrusive method of diagnosing illness in dairy cattle.

Table 3
Confusion matrix showing the classification of the odorous headspaces from the bacteria

Predicted class	True class	
	E. coli (180)	*S. aureus* (180)
E. coli (166)	166	0
S. aureus (188)	8	180
Unknown (6)	6	0

Table 4
Confusion matrix showing the optimal performance of bacterial phase classification using an af/a model. The accuracy of the classification is defined as $(2+154+133)/(14+162+182)$, i.e., 80.7%

Predicted class	True class of growth phase		
	Lag (14)	Log (162)	Static (182)
Lag phase (4)	2	2	0
Log phase (198)	12	154	32
Static phase (134)	0	1	133
Unknown (22)	0	5	17

3. Discussion

We have demonstrated that e-noses are able to identify unknown bacteria under laboratory conditions. This offers the possibility of using e-noses for lab-based, in-vitro diagnostics. The idea being that an e-nose could be used for the rapid screening of biological samples. For instance, the e-nose may be able predict a type of pathogen more rapidly (ca. 1 h) than existing semi-manual methods (ca. 1–2 days) and thus permit a more appropriate course of treatment. In this early stage of e-nose technology, it will be necessary to validate its predictions with existing, well-tested methods. However, should the lab-based e-nose technology prove reliable, then there is the very attractive option of moving the technology *near-patient*. For instance, Persaud and Travers [13] have reported upon the analysis of vaginal swabs to predict bacterial infection.

E-nose technology offers a number of significant advantages over other competing technologies. First, e-noses detect the major headspace volatiles and these may be related to the cell metabolism. In other words, the e-nose will only detect living cells and it is these viable cells that offer the threat to human health. Other techniques exist and can identify pathogens more specifically from protein sequences (e.g., biochips) but these will not indicate whether the microorganism is alive or dead.

The major competing technology to the lab-based e-nose for in-vitro diagnostics is probably mass spectrometry (MS) or gas chromatography–mass spectrometry (GC–MS). MS-based techniques can be used to identify specific molecules in, for example, the breath and these can then be associated with certain illnesses. Compounds such as dimethyl amine, acetone, hydrogen sulfide, and pentane have been reported in the literature to be associated with diabetes, uremia/kidney failure, lung carcinoma, liver disease and schizophrenia, respectively. However, the major advantages of an e-nose technology over MS-based methods is first that a chemical fingerprinting method may be more generic, but secondly and more importantly that

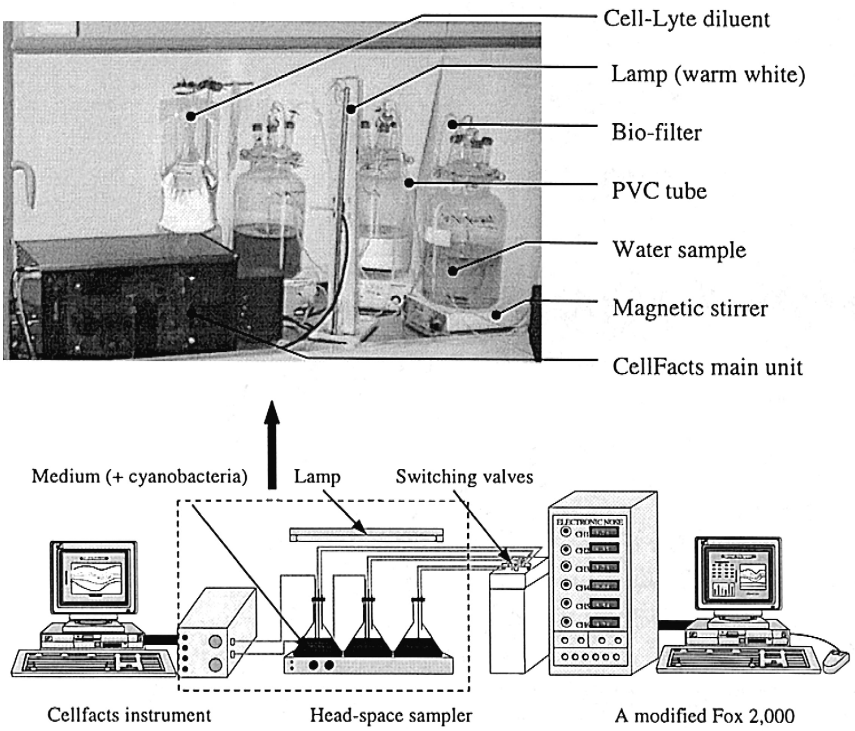

Fig. 4. Schematic diagram of the measurement system to collect data from water samples infected with cyanobacteria.

sensor-based e-noses can be fabricated at a much lower cost than MS-based analytical instrumentation. Thus the possibility of near-patient, low-cost units for at-patient or in-vivo diagnostics becomes a realistic option.

We anticipate that the next 3 years will lead to the commercial application of e-noses in lab-based in vitro medicinal diagnostics. We also believe that there are a number of technical challenges that need to be overcome before in-vivo diagnostics (e.g., mouth/breath) is realisable — such as the need to boost the sensitivity/selectivity of sensor-based e-noses to the key minor headspace volatiles (e.g., acetone on the breath), perhaps through the use of pre-concentrators, while in the presence of harmless major volatiles (e.g., TMA, H_2S and mercaptan that is normally associated with halitosis). The successful application of in-vivo e-noses for rapid screening would help to improve both our general health and welfare. Such e-noses would have significant financial value even if they were only able to diagnose illness with a limited success rate: they simply need to outperform statistically based clinical judgements.

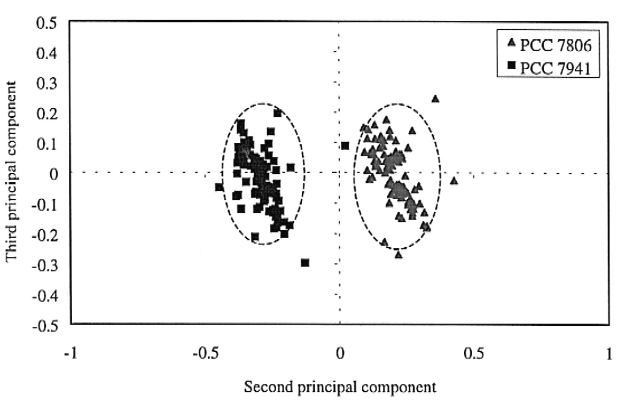

Fig. 5. Results of principal component analysis (PCA) plot of second and third principal components on anobacteria samples, PCC 7806 (toxic) and PCC 7941 (non-toxic). The original data are transformed by the normalised fractional difference model.

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Table 5
Confusion matrix for the prediction of state of health of the cows using only the sensitivity parameter K_i as the input to a one-output MLP

Diagnosis	Actual		
	Healthy	Sub-clinical	Clinical
Healthy	*7*	0	3
Sub-clinical	0	*0*	0
Clinical	5	3	*16*
Class size — 34	12	3	19

(Siloe Research Institute) for their contribution to one or more of the experiments reported here.

References

[1] H. Nanto, H. Sokooshi, T. Kawai, T. Usuda, Freshness detection of seafoods using ZnO thin film gas sensor, 10th Sens. Symp., Japan, 1991, pp. 195–198.
[2] R. Olafsson, E. Martinsdottir, G. Olafdottir, S.I. Sigfusson, J.W. Gardner, Monitoring of fish freshness using tin oxide sensors, in: J.W. Gardner, P.N. Bartlett (Eds.), Sensors and Sensory Systems for an Electronic Nose, Kluwer Academic Publishing, Dordrecht, Netherlands, 1992, pp. 257–272.
[3] M. Egashira, Y. Shimizu, Y. Takao, Fish freshness detection by semiconductor gas sensors, in: K. Kurihara, N. Suzuki, H. Ogawa (Eds.), Olfaction Taste XI, Sapporo, Japan, Springer-Verlag, Tokyo, Japan, 1994, pp. 715–719.
[4] M. Schweizer-Berberich, S. Vaihinger, W. Göpel, Characterisation of food freshness with sensor arrays, Sens. Actuators, B 18–19 (1994) 282–290.
[5] J. Olsson, T. Borjesson, J. Schnurer, Penicillium identification using an electronic nose, personal communication of conference abstract, 1995.
[6] F. Winquist, E.G. Hornsten, H. Sundgren, I. Lundström, Performance of an electronic nose for quality estimation of ground meat, Meas. Sci. Technol. 4 (1993) 1493–1500.
[7] B. Bourrounet, T. Talou, A. Gaset, Application of a multigas sensor device in the meat industry for boar-taint detection, Sens. Actuators, B 26–27 (1995) 250–254.
[8] T. Tan, Q. Lucas, L. Moy, J.W. Gardner, P.N. Bartlett, The electronic nose — a new instrument for sensing vapours, LC-GC Int. 8 (1995) 218–225.
[9] T. Nakamoto, A. Fukuda, T. Moriizumi, Perfume and flavour identification by odour-sensing system using quartz resonator array and neural network pattern recognition, Sens. Actuators, B 10 (1993) 85–90.
[10] J.R. Stetter, M.W. Findlay, K.M. Schroeder, C. Yue, W.R. Penrose, Quality classification of grain using a sensor array and pattern recognition, Anal. Chem. Acta 284 (1993) 1–11.
[11] A. Pisanelli, A.A. Qutob, P. Travers, S. Szyszko, K.C. Persaud, Applications of multi-array sensors to food industries, Life Chem. Rep. 11 (1994) 303–308.
[12] F. Winquist, H. Arwin, E. Lund, R. Forster, C. Day, I. Lundström, Screening of tomatoes by means of an electronic nose, 8th Int. Conf. Solid-State Sens. Actuators Eurosensors IX, Stockholm, Sweden, 1995, pp. 691–694.
[13] K.C. Persaud, P.J. Travers, Arrays of broad specificity films for sensing volatile chemicals, in: E. Kress-Rogers (Ed.), Handb. Biosens. Electron. Noses, CRC Press, Ohio, 1997, pp. 563–592.
[14] D.J. Strike, G. Fiaccabrino, N.F. de Rooij, M. Koudelka-hep, Headspace measurement of cheese samples using an electronic nose, Mater. Sci. Eng., C, in press.
[15] M. Craven, E.L. Hines, J.W. Gardner, P. Horgan, D. Morgan, I.A. Ene, Bacteria detection and classification using artificial neural networks in conjunction with an electronic nose, in: E.C. Ifeachor, K.G. Rosen (Eds.), Int. Conf. Neural Networks Expert Syst. Med. Healthcare, Bluestone Design, Plymouth, 1994.
[16] M. Craven, Bacteria classification with an electronic nose employing artificial neural networks, PhD thesis, University of Warwick, UK, 1997.
[17] T.D. Gibson, O. Prosser, J.N. Hulbert, R.W. Marshall, P. Corcoran, P. Lowery, E.A. Ruck-Keene, Detection and simultaneous identification of microorganisms from headspace samples using an electronic nose, Proc. Eurosensors X, Leuven, Belgium, September, 1996, pp. 1341–1344.
[18] R.J. Elliott-Martin, T.T. Mottram, J.W. Gardner, P.J. Hobbs, P.N. Bartlett, Preliminary investigation of breath sampling as a monitor of health in dairy cattle, J. Agric. Eng. Res. 67 (1997) 267–275.
[19] S. Chandiok, B.A. Crawley, B.A. Oppenheim, P.R. Chadwick, S. Higgins, K.C. Persaud, Screening for bacteria vaginosis: a novel application of artificial nose technology, J. Clin. Pathol. 50 (1997) 790–791.
[20] J.W. Gardner, M. Craven, C. Dow, E.L. Hines, The prediction of bacteria type and culture growth phase by an electronic nose with a multi-layer perceptron network, Meas. Sci. Technol. 9 (1997) 120–127.
[21] C. Mandenius, A. Hagman, F. Dunas, H. Sundgren, I. Lundström, A multisensor array for visualizing continuous state transitions in biopharmaceutical processes using principal component analysis, Biosens. Bioelectron. 13 (1998) 193–199.

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