CHAPTER 1

INTRODUCTION
1.1 Motivation

The first step to make earth a better place to live is to improve the quality of food provided to the consumer. Assurance of quality product is an important part of every company’s production. Due to competitiveness in food production, improper agricultural practices, poor hygiene and lack of preventive controls during food processing, misuse of chemicals, usage of contaminated raw materials, ingredients, water and improper storage, etc. made food unsafe for consumption [1]. According to Centers for Disease Control and Prevention (CDC), people often affected by foodborne illnesses due to consumption of food contaminated with disease-causing microbes. These types of contaminated food items are potentially life-threatening for young children, pregnant women and their fetuses, aged people, HIV / AIDS patients, cancer patients and people with weakened immune systems [2,3]. Common symptoms of foodborne illnesses include diarrhea, nausea, abdominal cramps, headache, dizziness and fever. In the developed / developing countries, surveillance of foodborne diseases is a fundamental component of food safety systems. According to the estimates of CDC in the year 2011, roughly 1 out of 6 Americans or 48 million people get sick, 128,000 are hospitalized and 3,000 die of foodborne illnesses [4]. Food poisoning bacteria’s can grow and multiply on food items more easily. Particularly perishable food items like meat, milk and poultry are considered as fast degradable foods [5]. Center for Science in the Public Interest (CSPI) field guide to meat and poultry food safety report for the period 1998 – 2010 has highlighted the foodborne illness outbreaks and classified them from highest to lowest risk levels (Fig. 1.1) [6]. With reference to this report risk levels of food illness have been decided based on the number of hospitalization and prolonged illness / death. According to this classification ground beef and beef meat
have been placed in the highest risk category. Hence researchers have started exploring
effective way to access the meat products.

![Risk Pyramid of Meat and Poultry Products]

Figure 1.1: A risk pyramid of meat and poultry products.

1.2 Meat quality

Highly perishable, fleshy foods like fish, meat and poultry have become an integral
part of human diet over many decades. World Health Organization (WHO) says the
consumption of meat products throughout the world has greatly risen in past few decades
because of their essential nutrients required for health. On the other hand, food safety is a
global issue and intake of spoiled meat causes several types of diseases. The main
contamination factors are toxic and harmful substances created by microorganisms
(bacteria, viruses and fungi) in raw meat. Especially *Escherichia coli* (*E. coli*) O157:H7
pathogen is responsible for high rate of hospitalizations. Since *E. coli* O157:H7 naturally
present in the intestinal of cattles, careless handling of meat during slaughtering lead to contamination of meat food. Nearly 90 percent of ground beefs were contaminated with *Salmonella*, *E. coli O157:H7* and *Listeria monocytogenes* [6,7]. In 1993, horrific foodborne-illness outbreak due to *E. coli O157:H7* in hamburgers sickened 700 people and killed 4 children in United States of America (USA) [8]. After that incident, USA government declared that meat and poultry industries are to be monitored under Hazard Analysis and Critical Control Point (HACCP) by the United States Department of Agriculture (USDA) Food Safety and Inspection Service (FSIS). Shortly thereafter meat industries have made changes in animal production, slaughtering and processing methods to reduce the risk of pathogens. Several precautionary methods can reduce the bacterial contamination but still cannot be cleared completely. Since, the presence of microbes are inevitable, their concentration in the meat should be monitored. To monitor their presence and the quality of meat, simple and rapid techniques are best suited in many cases.

### 1.3 Meat quality testing methods

Meat can be contaminated by bacteria which can spread through water, air, soil, workers and equipment involved during the manufacturing process. Traditionally, various techniques like salting, freezing, canning, smoking, modified atmosphere packaging, etc., have been used to preserve meat. Quality assessment of meat during processing, packaging, transporting, storing, displaying and cooking is necessary on regular basis to ensure the quality. To prevent wastage and food borne illnesses, various testing methods were employed to certify the quality of meat. In general meat quality can be assessed either by examining the structure (texture, tenderness, flavour, juiciness, and colour) or by detecting
the microorganisms and their count or by detecting the gases / volatile organic compounds (VOCs) generated by these microorganisms [9,10]. The techniques that are available to examine the structure of meat are Warner–Bratzler shear force test [11], ultrasonic elastography [12], Raman spectroscopy [13], colorimetry [14], fluorescence spectroscopy [15], optical spectroscopy [16], electron microscopy [17] and magnetic resonance elastography [14]. The quality analysis of meat by examining its structure is time consuming, complex, destructive and expensive. From the point of rapid quality discrimination with greater accuracy, sensory evolution is found to be the best method from literature. The practical application of human nose as a smell assessment instrument is severely limited by the fact that our sense of smell is subjective, gets tired easily, and is therefore difficult to use. Consequently, there is a considerable need for an instrument that could mimic the human sense of smell and its feasibility in industrial applications.

1.4 **Electronic nose**

Generally accepted definition of an electronic nose (e-nose) is an instrument, comprises an array of heterogeneous electrochemical gas sensors with partial specificity and a pattern recognition system [18,19]. However, in more recent years, the term e-nose has been used in a broader sense to refer gas sensors that measure the ambient gas atmosphere based on the general principle that changes in the gaseous atmosphere alter the sensor properties in a characteristic way. A variety of sensor types have been developed in which three types of materials are commonly used: metal oxides, conducting polymer composites and intrinsically conducting polymers. Apart from conductive sensors, gas detection has also been done using optical sensors, surface acoustic wave sensors, gas
sensitive field effect transistors and quartz microbalance (QMB) sensors. Micro-electro-mechanical systems (MEMS) plus nanotechnologies are the most promising emerging technologies in the field of gas sensors. The term e-nose has also been used to characterize systems where ultra-fast gas chromatography or mass spectrometry is employed in the detection process. Once the data from sensors array are collected, the e-nose system requires a suitable post processing procedure to analyze and classify the data. Pre-processing of multivariate signals in sensor arrays represents an essential part of the measuring system. Data processing techniques used in post processing of pattern recognition routines include principal component analysis (PCA), linear discriminate analysis (LDA), partial least squares (PLS), functional discriminate analysis (FDA), cluster analysis (CA), fuzzy logic or artificial neural network (ANN) such as probabilistic neural network (PNN). Among these techniques, PCA, PLS, LDA, FDA and CA are based on a

Figure 1.2: Illustration of basic analogy between human olfaction and e-nose [67].
linear approach while fuzzy logic, ANN and PNN are regarded as nonlinear methods [20].

Fig. 1.2 illustrates a basic analogy between artificial and biological olfaction, where array
of sensors fill the role of olfactory sensory receptor cells, while microcontroller-based
pattern-recognition algorithms replace the neural circuitry of the brain.

E-nose instruments are attractive for a number of significant features: the relatively
fast assessment of headspace, a quantitative representation or signature of a gas and low
cost sensors which can be easily integrated in current production processes. Despite these
features, there are still relatively few applications of e-noses adopted in industry. This could
be attributed to difficulties in robustness, selectivity and reproducibility of the sensors and
to the need for pattern recognition algorithms which can cope with the complex signal
analysis. Nonetheless, the use of e-noses is rapidly expanding and there have been notable
achievements relevant for the food industry, particularly in the past few years. Furthermore,
this progress coincides with an increased understanding of the biological mechanisms
behind the human olfactory system. Specifically, we now have a greater understanding not
only of the genetics behind the olfactory receptors but also of the relationships between an
odorant’s molecular property and the quality of an odour. Today, several e-noses are
commercially available in the market. These e-noses are often promoted as generic devices
and suitable for a range of applications. Table 1.1 provides a list of some of the common
commercially available e-nose models.
Table 1.1: A list of some of the common commercially available e-nose models.

<table>
<thead>
<tr>
<th>Model</th>
<th>Number of sensors</th>
<th>Technology</th>
<th>Manufacturer</th>
<th>Country</th>
</tr>
</thead>
<tbody>
<tr>
<td>i-Pen, PEN2, PEN3</td>
<td>6</td>
<td>MOS Sensor</td>
<td>Airsense Analytics</td>
<td>Germany</td>
</tr>
<tr>
<td>QCS</td>
<td>2</td>
<td>MOS Sensor</td>
<td>Gerstel GmbH &amp; Co. KG</td>
<td>Germany</td>
</tr>
<tr>
<td>Artinose</td>
<td>38</td>
<td>MOS Sensor</td>
<td>Sysca AG</td>
<td>Germany</td>
</tr>
<tr>
<td>FF2</td>
<td>6</td>
<td>MOS Sensor</td>
<td>RST Rostock System-Technik GmbH</td>
<td>Germany</td>
</tr>
<tr>
<td>GFD1</td>
<td>6</td>
<td>MOS Sensor</td>
<td></td>
<td>Germany</td>
</tr>
<tr>
<td>FOX 2000, 3000 &amp; 4000</td>
<td>6, 12 &amp; 18</td>
<td>MOS Sensor</td>
<td>Alpha MOS</td>
<td>France</td>
</tr>
<tr>
<td>Promethus.</td>
<td>18</td>
<td>MOS Sensor</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Air quality module</td>
<td>2</td>
<td>MOS Sensor</td>
<td>Applied Sensor</td>
<td>Sweden</td>
</tr>
<tr>
<td>EOS 835, Ambiente</td>
<td>6</td>
<td>MOS Sensor</td>
<td>Sacmi</td>
<td>Italy</td>
</tr>
<tr>
<td>Bloodhound ST 214</td>
<td>14</td>
<td>Conducting polymers</td>
<td>Scensive Technologies</td>
<td>UK</td>
</tr>
<tr>
<td>Aromascan A32S</td>
<td>32</td>
<td>Conducting polymers</td>
<td>Osmetech Plc</td>
<td>USA</td>
</tr>
<tr>
<td>Cyranose 320</td>
<td>32</td>
<td>Conducting polymers</td>
<td>Intelligent Optical Systems, Inc.</td>
<td>USA</td>
</tr>
</tbody>
</table>

1.5 Application areas in food

E-nose finds its applications in various fields like environmental monitoring, food industries, medicine, automobile industry and so on [21,22]. Since 1993, the number of publications in the area of electronic olfaction is more than 12,000 articles. Approximately 5000 publications mainly emphasis the implementation of sensors in quantifying the
quality of fish, meat, milk, wine, coffee and tea. This demonstrates that food applications are central to electronic olfaction and nearly half of the publications are in this area. Within each application in the food industry, research contributions have focused on the detection of a variety of aspects such as freshness, adulteration, off-flavours and bacteria detection. Table 1.2 presents a summary of the applications of e-nose in different food matrices.

Table 1.2: Summary of the applications of e-nose in different food matrices.

<table>
<thead>
<tr>
<th>Application</th>
<th>Sensing element</th>
<th>E-nose model</th>
<th>Number of sensors</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rancidity of two different kinds of milk (ultrahigh temperature processed and pasteurized)</td>
<td>MOS (SnO₂)</td>
<td>Lab Made</td>
<td>5</td>
<td>[23]</td>
</tr>
<tr>
<td>To detect unspoiled and spoiled milk</td>
<td>Conducting polymer</td>
<td>Bloodhound Sensors Ltd. UK (BH-114)</td>
<td>14</td>
<td>[24]</td>
</tr>
<tr>
<td>To discriminate different milk flavourings</td>
<td>MOS</td>
<td>Alpha MOS, France (Fox 4000)</td>
<td>18</td>
<td>[25]</td>
</tr>
<tr>
<td>Identification of adulterated milk</td>
<td>MOS</td>
<td>Airsense Analytics, Germany (PEN 2)</td>
<td>10</td>
<td>[26]</td>
</tr>
<tr>
<td>Identification of odours (milk, rancid milk and yoghurt)</td>
<td>MOS</td>
<td>Figaro Engineering Inc., Japan (TGS 2620, 2610 and 2600)</td>
<td>5</td>
<td>[27]</td>
</tr>
<tr>
<td>Differentiate the milk collected from healthy and mastitic disease affected cows</td>
<td>MOSFET + MOS</td>
<td>Applied Sensor, Sweden (3320)</td>
<td>22</td>
<td>[28]</td>
</tr>
<tr>
<td>Seasonal changes in milk powder</td>
<td>Conducting polymer</td>
<td>OSMETECH PLC, England</td>
<td>28</td>
<td>[29]</td>
</tr>
<tr>
<td>Bacterial contamination in milk</td>
<td>QCM</td>
<td>SES Piezo Ltd., England</td>
<td>6</td>
<td>[30]</td>
</tr>
<tr>
<td>Detect different off-flavours in wines of different origin</td>
<td>MOS</td>
<td>Alpha MOS, France (Fox 4000)</td>
<td>18</td>
<td>[31]</td>
</tr>
<tr>
<td>Task</td>
<td>MOS</td>
<td>Sensor Details</td>
<td>Score</td>
<td>Reference</td>
</tr>
<tr>
<td>----------------------------------------------------------------------</td>
<td>--------------</td>
<td>--------------------------------------------------------------------------------</td>
<td>-------</td>
<td>-----------</td>
</tr>
<tr>
<td>Prediction of Italian red wine</td>
<td>MOS</td>
<td>Airsense Analytics, Germany (PEN 2)</td>
<td>10</td>
<td>[32]</td>
</tr>
<tr>
<td>Classification of four types of red wines</td>
<td>MOS (SnO₂)</td>
<td>Lab Made</td>
<td>16</td>
<td>[33]</td>
</tr>
<tr>
<td>Detection of aroma from green tea at different storage times</td>
<td>MOS</td>
<td>Airsense Analytics, Germany (PEN 2)</td>
<td>10</td>
<td>[34]</td>
</tr>
<tr>
<td>Identification of optimum fermentation time for black tea</td>
<td>MOS</td>
<td>Figaro Engineering Inc., Japan (TGS-832, 823, 831, 816, 2600, 2610, 2611 and 2620)</td>
<td>8</td>
<td>[35]</td>
</tr>
<tr>
<td>Quality evaluation of black tea</td>
<td>MOS</td>
<td>Figaro Engineering Inc., Japan (TGS 832, 823, 2600, 2610, and 2611)</td>
<td>5</td>
<td>[36]</td>
</tr>
<tr>
<td>Quality evaluation of black tea</td>
<td>MOS</td>
<td>Figaro Engineering Inc., Japan (TGS 832, 823, 2600, 2610 and 2611)</td>
<td>5</td>
<td>[37]</td>
</tr>
<tr>
<td>Classification of black tea</td>
<td>MOS</td>
<td>Figaro Engineering Inc., Japan (TGS 816, 823, 831, 832, 2600, 2610, 2611 and 2620)</td>
<td>8</td>
<td>[38]</td>
</tr>
<tr>
<td>Discriminating different grades of green teas</td>
<td>MOS</td>
<td>Airsense Analytics, Germany (PEN 2)</td>
<td>10</td>
<td>[39]</td>
</tr>
<tr>
<td>Distinguish the Japanese green teas with different content of coumarin</td>
<td>MOS</td>
<td>Fragrance &amp; Flavor Analyzer, Shimadzu, Japan (FF-2A)</td>
<td>10</td>
<td>[40]</td>
</tr>
<tr>
<td>Discrimination of tea with different qualities</td>
<td>MOS</td>
<td>Figaro Engineering Inc., Japan (TGS 880, 826, 825 and 822)</td>
<td>4</td>
<td>[41]</td>
</tr>
<tr>
<td>Classification of different brands of Espresso coffee</td>
<td>MOS (SnO₂)</td>
<td>Lab Made</td>
<td>4</td>
<td>[42]</td>
</tr>
<tr>
<td>Classification of different coffee</td>
<td>MOS</td>
<td>Figaro Engineering Inc., Japan (TGS 800, 815, 816, 821, 823, 824, 825, 830, 831, 842, 880, 881, 882, 883)</td>
<td>14</td>
<td>[43]</td>
</tr>
<tr>
<td>Application</td>
<td>Sensor Type</td>
<td>Manufacturer</td>
<td>Shelf Life</td>
<td>Reference</td>
</tr>
<tr>
<td>-------------------------------------------------</td>
<td>-------------</td>
<td>-------------------------------------------------------------------------------</td>
<td>------------</td>
<td>-----------</td>
</tr>
<tr>
<td>Analysis of Colombian coffee samples</td>
<td>MOS</td>
<td>Figaro Engineering Inc., Japan and FIS Inc., Japan (SP-12A, SP 31, TGS 813, TGS 842, SP-AQ3, TGS 823, ST-31 and TGS 800)</td>
<td>8</td>
<td>[44]</td>
</tr>
<tr>
<td>Shelf-life determination of red meat</td>
<td>MOS</td>
<td>Figaro Engineering Inc., Japan (TGS 815, 821, 822, 824, 825 and 842)</td>
<td>6</td>
<td>[45]</td>
</tr>
<tr>
<td>Discrimination of boar tainted samples of different meat parts</td>
<td>MOS</td>
<td>Alpha MOS France (Fox 4000)</td>
<td>18</td>
<td>[46]</td>
</tr>
<tr>
<td>Quality detection of porcine meat loaf</td>
<td>MOS</td>
<td>Danish Odour Sensor System (DOSS)</td>
<td>6</td>
<td>[47]</td>
</tr>
<tr>
<td>Detection of boar taint</td>
<td>MOS</td>
<td>Figaro Engineering Inc., Japan (TGS 825, 882, 824, 822, 800)</td>
<td>5</td>
<td>[48]</td>
</tr>
<tr>
<td>Spoilage classification of red meat</td>
<td>MOS</td>
<td>Figaro Engineering Inc., Japan (TGS 823, 825, 826, 831, 832, 882)</td>
<td>6</td>
<td>[49]</td>
</tr>
<tr>
<td>Meat spoilage markers detection</td>
<td>MOS (ZnO)</td>
<td>Lab Made</td>
<td>3</td>
<td>[50]</td>
</tr>
<tr>
<td>Discrimination of microbial population</td>
<td>MOS</td>
<td>Figaro Engineering Inc., Japan (TGS 812, 822, 880, 2602, 2611 and 2611)</td>
<td>6</td>
<td>[51]</td>
</tr>
<tr>
<td>Quality identification of rapid fermented fish</td>
<td>MOS</td>
<td>Alpha MOS, France (Fox 4000)</td>
<td>18</td>
<td>[52]</td>
</tr>
</tbody>
</table>
1.6 Meat quality biomarkers

Meat export industries have taken several steps to prevent the decomposition by reducing the microbial spoilage factor. Spectroscopy, culture based and imaging techniques are used to test the freshness levels of meat. But these techniques are having their own advantages and limitations [13]. Sensory analysis is considered to be the best tool to determine the meat freshness in the industries and consumer markets [53]. The schematic view of the developed e-nose using readymade sensors and traditional culture-based method are shown in Fig. 1.3. The first protocol of assessing meat quality using e-nose is to identify the biomarkers. Biomarkers are metabolic by-products (chemicals / gases) produced by dominant spoilage microorganisms as a result of their growth in food. Sensing the concentration levels of these biomarkers [54–56] using e-nose is found to be

![Figure 1.3: Schematic of the proposed method and traditional culture-based method.](image)

<table>
<thead>
<tr>
<th>Proposed Method</th>
<th>Traditional Method</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image" alt="Image" /></td>
<td><img src="image" alt="Image" /></td>
</tr>
</tbody>
</table>

Handy Equipment – Rapid Output (< 1 minute)

Laborious Process – Slow Output (~ 1-2 Days)
one of the best and promising solutions to analyse the quality of meat [57,58]. Fatty acids, amino acids, proteins, fat content, flavour, colour and texture of the meat depends upon the farming systems, control of growth rate, feeding behaviour, nutrition intake and freshness. Production of VOCs and non VOC from meat food primarily due to lipid and / or protein oxidation [59]. Fatty acids profile, aldehydes (acetaldehyde, hexanal, 2-methyl-1-butanal, nonanal, 2, 4-heptadial), ketones (2-octanone, 2-decanone, 2-propanone), trimethylamine, ammonia, volatile organic compounds (1-butanol, 1-penten-3-ol) sulfur compounds (hydrogen sulfide) are the major compounds identified as the biomarkers of meat spoilage levels [10,60–62]. There are thousands of volatile compounds identified on headspace of meat but not all of them are important. According to literature, ammonia, acetaldehyde and hydrogen sulfide play a key role in overall aroma flavour. Production of ammonia and acetaldehyde in meat is due to catabolism of amino acids by bacteria [9,63,64]. Also the conversion of myoglobin to sulfmyoglobin results in the production of hydrogen sulfide [65,66]. Hence, sensing these three biomarkers will help to effectively assess the quality of meat food.

1.7 Focus of current research

Metal oxide semiconductor based sensors are found to be suitable candidates due to their high sensitivity and compactness for detecting VOCs / gases at various concentrations. Among various metal oxide semiconductors, zinc oxide (ZnO) is found to be one of the most promising materials for gas sensing applications due to its excellent response towards VOCs / gases, high chemical stability, suitability for doping, non-toxicity, availability and low cost. In particular, ZnO with different nanostructured
morphologies has been preferred for sensing applications because of its high surface to volume ratio, grain dimension comparable to the space charge region, tunable sensitivity and superior stability. ZnO thin films were deposited by several methods, but spray pyrolysis technique was preferred in this work owing to its advantages such as simplicity, large productivity, cost effectiveness and environmentally safe since water was used as solvent and requirement of no vacuum.

Majority of the commercially available gas sensors have been designed with the micro heater to enhance the response of the sensing element at elevated operating temperatures. Because of switching between heating and normal atmospheric conditions, structural transformation, phase separation, grain growth of the sensing element as well as degradation of contacts lead to poor stability and decreased life time. In this scenario, detection of target gases at room temperature would be one of the best solutions to overcome the problem of degradation of the sensing element and also such sensors can be employed for the in situ detection of gases like ammonia at low temperatures and explosive environments. Selectivity is another major challenge of metal oxide sensors, which limits their utilization in mixed gas / vapour atmosphere. Hence in this work, highly sensitive and selective room temperature sensors for the detection of meat quality biomarkers such as ammonia, acetaldehyde and hydrogen sulfide have been developed. The developed gas / chemical sensors can be employed as a part the sensor array in an electronic nose for meat quality assessment.
1.8 Objectives

The overall objective of this thesis work is to develop sensing elements using ZnO nanostructures to detect meat quality biomarkers namely ammonia, acetaldehyde and hydrogen sulphide with enhanced sensitivity, selectivity, stability, response and recovery time. The following specific aims were formulated and achieved towards the proposed objective.

Specific Aim #1

Development of highly sensitive and selective room temperature ammonia sensor based on ZnO nanospheres

Specific Aim #2

Development of highly sensitive and selective room temperature acetaldehyde sensor based on ZnO nanoplatelets

Specific Aim #3

Development of highly sensitive and selective room temperature hydrogen sulfide sensor based on ZnO nanorods

Specific Aim #4

Packaging of sensing elements on printed circuit boards
1.9 Thesis organization

This thesis consists of seven chapters as follows:

Chapter 1 presented an overview of the importance of meat quality and its assessment techniques, application of e-nose in meat assessment, focus of current research, goals and objective of this research work.

Chapter 2 describes the literature review on the basic characteristics of gas sensors, principle of operation, role of morphology, dopants, annealing and also highlights the results of ZnO sensors available in the literature with particular focus on spray pyrolysis technique.

In addition, materials and methods, characterization techniques employed in this work have been presented.

Chapter 3 focusses on the development of room temperature ammonia sensor using the spray deposited ZnO films with spherical nanograin structure. This chapter includes the structural, morphological, optical, electrical and room temperature ammonia sensing studied of the developed ZnO sensing element.

Chapter 4 explains the growth of template free ZnO nanoplatelets using spray pyrolysis technique. This chapter emphasises the role of surface morphology, annealing temperature in the acetaldehyde sensing performances of ZnO film.

Chapter 5 discusses the successful synthesis of ZnO nanorods of various packing densities without any template. Also this investigation revealed the hydrogen sulfide sensing behaviour of ZnO nanorods at room temperature.

Chapter 6 depicts the sensor packaging schemes and discusses the key features of the developed sensors.

Chapter 7 summarizes the entire results and concludes with future directions.
1.10 References


