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Recent advances in wearable sensors for animal health management



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ABSTRACT

Biosensors, as an application for animal health management, are an emerging market that is quickly gaining recognition in the global market. Globally, a number of sensors being produced for animal health management are at various stages of commercialization. Some technologies for producing an accurate health status and disease diagnosis are applicable only for humans, with few modifications or testing in animal models. Now, these innovative technologies are being considered for their future use in livestock development and welfare. Precision livestock farming techniques, which include a wide span of technologies, are being applied, along with advanced technologies like microfluidics, sound analyzers, image-detection techniques, sweat and salivary sensing, serodiagnosis, and others. However, there is a need to integrate all the available sensors and create an efficient online monitoring system so that animal health status can be monitored in real time, without delay. This review paper discusses the scope of different wearable technologies for animals, nano biosensors and advanced molecular biology diagnostic techniques for the detection of various infectious diseases of cattle, along with the efforts to enlist and compare these technologies with respect to their drawbacks and advantages in the domain of animal health management. The paper considers all recent developments in the field of biosensors and their applications for animal health to provide insight regarding the appropriate approach to be used in the future of enhanced animal welfare.

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1. Introduction

The use of biosensors and wearable technologies is becoming increasingly important for animal health management. These devices, if built precisely and used correctly, can provide timely diagnosis of diseases in animals, eventually decreasing economic losses. Such devices are particularly useful for dairy cattle and poultry farms. Instead of relying solely on farmers' senses and knowledge, on-site sensors can provide reliable data about the physical condition of the animals. Due to the superior performance of wearable technologies and sensors, they can make a breakthrough in livestock development, and promises to become one the most impactful and practicable technology in the animal health market. New wearable technologies are being customized to meet the needs of animals, pets and livestock. Products such as medication patches, tracking collars, and electronic saddle optimization are being purchased at higher rates [1] and harnessed for the healthier upbringing of farm animals. These wearable technologies are multifunctional and efficient, allowing animal owners to do more in less time. Global growth of this sector in the next ten years has been predicted to soar from \$0.91 billion to \$2.6 billion [1].

Sensors and wearable technologies can be implanted on animals to detect their sweat constituents [2–4], measure body temperature [5–7], observe behavior and movement [8,9], detect stress [10], analyze sound [11–16], detect pH [17], prevent disease [18], detect analytes and detect presence of viruses and pathogens [19–23]. Wearable sensors help farmers catch disease early, and thereby prevent deaths of animals. Farmers can also cull diseased animals in time to prevent the spread of disease in whole cattle herds through prediction.

Apart from collecting useful data regarding animal health, general farm monitoring can also be made easier and more reliable by using biosensors integrated with cellphones and handheld devices instead of conventional methods, such as writing notes, keeping a farm diary, or using simple equipment without data-sharing functions. A number of systems have been developed on cellphones and handheld devices to reduce the effort of recording data manually [24]. Solar-powered receivers mounted on livestock can collect data that is transmitted to a central server. The final data can easily be viewed on a custom dashboard or office computer, which makes this technology very convenient for farmers.

A biosensing device that attaches to ears to measure the body temperature of animals now costs \$100,000 for 10,000 cattle. Commercially available biosensor collars are also being used in cows for detection of estrus period [25–27]. An innovative robotic grazing system uses electronic leg bands that interact with sensors mounted on the animal to record data on its feeding and milking behavior and pattern [28].

It's a big challenge to provide good quality, safe meat to meet the increasing global demand for meat and poultry products. With rising demand comes growing concerns relating to animal health [29]. Devices that can be integrated inside the body of animal, patched under its skin, or remain in its stomach give animal owner's useful information regarding their behavior and medical conditions. These electronic devices are expected to be used for the medical treatment of animals,

detection of heating and cooling needs, iontophoretic drug delivery, and even conservation of wild species [1].

Another important use of biosensors is antibiotic detection. With the unhampered and frequent use of antibiotics in the animal industry, antibiotic resistance has become a major threat for farmers. Ecological instability is caused by the uncontrolled use of sub-therapeutic antibiotics in concentrated animal feeding operations (CAFOs), which in turn causes antibiotic resistance in animals. There is a dire need for farmers to switch to alternatives to avoid animals becoming immune to antibiotic treatment. The amount of antibiotics administered in the blood serum and muscles of farm animals should be kept in a certain range, and there should be a proper system to detect the antibiotic levels in the animal body. It is nearly impossible to put a ban on the use of antibiotics in the livestock health management, since antibiotics help cure the most common ailments, like enteric and respiratory infections. The use of antibiotics in sub-therapeutic concentrations for increasing development and growth of farm animals is also well recognized. To address this prevailing issue, the European Union set up a standard to prevent the antibiotic resistance. This principle, which has been suggested as precautionary measure, focuses on banning certain antimicrobial growth promoters. Maximum Residue Limits (MRLs) have been set up for those antibiotics that are still allowed to be administered in animals in the United States and European countries. MRL is that amount of pharmacologically active substances, and their derived metabolites, which is legally acceptable. Biosensors have been identified as being helpful in this regard; they can easily detect antibiotic levels and warn the farmer if the antibiotics level exceeds a maximum range [19].

The international market for wearable technology for animals is expected to grow from around \$1 billion to \$2.5 billion in the next decade, increasing more than 2.5 times [1]. The highest percentage of manufacturers of this unique technology is in China, which is providing these products at a very cheap price, followed by the USA.

A significant amount of money is spent every year on agricultural research and animal health management. However, this does not necessarily translate to better productivity or increased health of animals. More often than not, the funding is aimed to provide newer solutions to the problems, rather than bridging the gap between research and industry. Banhazi and Black [30] have suggested that a rigorous procedure be carried out to ensure that agricultural practices are correct and consistent in accordance with the current knowledge and research findings [30]. This ambitious standard can only be accomplished by integrating data measurements and data acquisition systems through novel biosensing technologies.

To meet the current and emerging challenges of farmed animal disease surveillance, diagnostics and control, it is imperative that a paradigm shift occurs in how diseases are identified. This shift involves replacing the shipping samples from farms to labs with rapid diagnosis on the farm itself. The world organization for animal health (OIE) has warned that the zoonotic diseases from farmed animals can have devastating impacts on public health if there is spill over from the farmed animal reservoir, and the livestock industry is under heavy pressure to improve its biosecurity protocols and enhance animal traceability and

welfare. Hence, the livestock industries and watch dog food safety inspection agencies are seeking new tools and technologies, to enable rapid, real-time and on-farm monitoring of diseases and record keeping.

Biosensing technologies provides promise to improve the performance, cost, and productivity in the area of disease management in livestock. Development and deployment of reliable, rapid tests will allow earlier and more specific treatment of diseases, potentially resulting in reduced antimicrobial usage and improved animal welfare. In addition, the biosensors and sensing technologies to alert producers of diseases even before the disease occurs will be a novelty and could form management components of the integrated farm inspection and proAction model. The opportunity to detect diseases and factors concerning milk production through the use of technologies will establish risk mitigation in on-farm operations and thereby will enhance the animal care and biosecurity components of proAction.

The integration of novel diagnostic and disease detection systems using biosensors would keep livestock and agricultural industry one step ahead of invisible diseases through satellites and smartphones. Smart and precision livestock farming and animal health management will continue to grow in importance to meet the increasing demand for food and ensure sustainability in farming. The biosensing technologies with the advances in the internet of things (IoT) paradigm will promote rapid, on-farm and real-time monitoring of farmed animal diseases. The real-time dissemination of data collected from the farms through these biosensors will have value beyond the farm as well; allowing food manufacturing stakeholders' access to this information that will prove essential to the social license issues facing our agricultural sector and will be a key to our continued global competitiveness.

Early detection of diseases using biosensors allows for shifting of the epidemiological curve to the left by enabling rapid response, reducing the spread of the disease and associated production, social and economic consequences. Reducing the time to obtain results in diagnosing infectious disease biomarkers on-farm in a real time fashion will provide an early warning system for smart livestock health management.

The purpose of this review article is to describe, compare and analyze various wearable technologies that have been developed recently towards providing solutions for farmed animal health management.

2. Biosensors and underlying technologies

The ability to quickly, accurately and reliably detect the presence or absence of biomarkers or specific chemicals can be a matter of life or death of the farmed animals. Monitoring of glucose or proteins or enzymes in the bloodstream, testing for harmful compounds such as metals or antibiotic residues in animals, and early warning of the biological and chemical agents in the livestock animal health sector requires sensitive and reliable sensing devices. While the demand for real-time detection of diseases using the sensors and devices are ever more urgent, the capability of several relevant enabling technologies to build the sensing devices is also unprecedented. Bionanotechnology and microelectronics made it possible to fabricate transistors smaller than 100 nm and to integrate several hundreds of them into a functional circuit on a small chip. Rapid progresses in nanofabrication has also offered novel enabling technologies.

2.1. Biosensors

A variety of methods are used to detect the level of antibiotics in the body to avoid health hazards. Biosensors are the most prevalent method. The mechanism and configuration of biosensors is very simple and easy to understand, and provides fast, accurate detection of antibiotics. Biosensors work with the help of a recognition element and a transducing device. The recognition element works on the mechanism of affinity-pairing, such as enzyme/substrate and antibody/antigen receptors (Fig. 2). The transducer detects any contact between such pairs by producing detectable electrical signals in response to biological activity,

which is later analyzed. The use of biosensors is limited in the field currently mainly because the biological sensing element is affected by different factors, including environmental factors and type of molecules. Moreover, the size of transducer can also affect the efficiency and functioning of a biosensor [19].

2.1.1. Antibiotic detection

Fig. 1 illustrates the distribution of analytical methods used for determining antibiotic levels in food (Figs. 2–5).

2.1.2. Microfluidics

Another technology which is becoming widely used is microfluidics, which makes the rapid detection of analytes possible. In different disciplines, including food safety, the detection of analytes is of primary importance, and this technology has proved to be advantageous in this regard. Microfluidics technology is able to utilize small samples, which can be quickly detected and lead to less reagent wastage. Furthermore, the utilization of microfluidics technology in Point of Care (POC) operations has been successful, as this substantially reduces the risk of cross contamination [31]. The use of paper-based and thermoplastic chips has revolutionized the development of disease diagnostic platforms. Paper-based chips exclude the need for preprocessing samples. On the other hand, thermoplastic chips are feasible because they are disposable and cost effective [19].

2.1.3. Fluorescence resonance energy transfer (FRET)

Integration of microfluidics and fluorescent labels ensures not only the requirement of minimum sample volume, but also an enhancement in sensitivity by successfully reducing the background signal noise. Microfluidics technology also aids in minimizing the signal-to-noise ratio, and in reducing the noises during the measurement of Raman scattering and Rayleigh stray light signals during biomarkers or analyte detection. The chip material for microfluidics technology should be suitable for microscopy, and possess characteristics like being non-adsorbent to the molecules. DNA analysis can also be done by the use of this versatile technology by incorporating fluorescence resonance energy transfer (FRET).

2.1.4. Quantum dots

Features of microfluidics, such as allowing spatial and temporal resolution and easy differentiation between non-hybridizing and hybridizing oligomers of DNA, give it an advantage over previously used technologies. However, since issues of pH sensitivity and photo bleaching are common in FRET-based techniques, quantum dots (QD) are utilized to provide stability. QD has an extensive emission wavelength, which can be adjusted by changing their size and composition with other nanomaterials [19].

2.1.5. Surface Plasmon Resonance technology (SPR)

SPR technology makes the future of biosensors very promising. About 20 commercial standard SPR platforms are currently present on the market. The application of gold nanoparticles for detecting antibiotics in food residues has caught the attention of the food industry. Recently, nanoparticles have been shown to amplify SPR signals. Gold nanoparticles upon combination with screen-printed electrodes can produce versatile and suitable electrochemical properties. The physical and chemical properties of these particles, such as efficient mass transport and enhanced surface area, are desirable due to the high surface to volume ratio. Gold nanoparticles have found their application in antibiotic detection due to a fast binding rate with the biomolecules and low toxicity of these versatile particles. Using Solid Phase Extraction (SPE) combined with Self Assembled Monolayer (SAM) cysteine (Cys) on gold nanoparticles also raises the selectivity of this technique and boosts the sensitivity, by increasing efficiency.

Using impedimetric immunosensors is another highly sensitive and fast way to detect biomarkers and analytes of interest. Integrating the

soluble gel-derived silica-based material with these immunosensors ensures encapsulation of biorecognition elements with adequate mechanical stability.

The most recent development in biosensing technology is the introduction of an SPR device for portable detection of antibiotics. This approach is very practical and gives highly sensitive results for antibiotic detection in chicken muscle/blood serum in slaughterhouses.

However, there is still a large growth potential for technology that detects larger families of antibiotics more easily and quickly [19], and applied SPR to the detection of catalase in milk samples [32,33].

2.1.6. Hybrid technologies

The combinations of QD, FRET and microfluidics ensure that the signal is amplified during DNA analysis and the molecular size is accurately detected. The combination of gold nanoparticles and microfluidics is reported to be 150 times more sensitive than the traditional ELISA method in the detection of interleukin-2. Digital microfluidics with SPR is applicable for industrial use due to its astonishingly accurate screening. This technology is convenient, since it does not require labelling prior to analysis [19].

There are, however, some disadvantages associated with this technology, such as non-specific adsorption and the difficulty of functionalizing metal surfaces. Microfluidics can also be integrated with Surface-enhanced Raman Spectroscopy or Surface-enhanced Raman Scattering (SERS). This technique increases Raman scattering by molecules adsorbed on rough metal surfaces. The unique combination of microfluidics and SERS results in precise, reproducible results and consistent mixing conditions. The drawback of this combination is the unfeasible size of Raman instruments for detection. Microfluidics and SERS combination is being used for analyte detection for homeland security, and has significantly aided the successful recognition of cells leading to cancer [19].

2.2. Sweat analyzers

Analyzing sweat can relay useful information about an individual animal's health [34–39]. Wearable sweat analyzers have not yet been made commercial, mainly because of the size constraints of the equipment. However, low-cost robust designs have been developed in laboratories [40,41]. Methods for collecting sweat include using an electrical current to drive a chemical stimulant into the skin—iontophoresis—but there is a need for methods that not only collect but also analyze and monitor sweat throughout the day or as required [3]. Recent developments made in sweat analyzers aim to restrict the size of the system so it is wearable and easy to handle (Table 1). Real-time sweat monitoring of sodium by disposable potentiometric strips integrated with microfluidic chips has been developed; it is connected to a mini wireless system to detect sodium levels in sweat [42]. Monitoring a number of electrolytes simultaneously is more useful; hence, the system developed by Gao et al. conveys levels of sodium, potassium, lactate, glucose and skin temperature simultaneously. Integrated Bluetooth technology enables sharing and monitoring of the measured data [43]. Biomonitoring of sweat in animals has great potential for animal health because of its non-invasive nature. The amount of metals can also be detected by sweat analyzers [44]. If such a technology is introduced on farms, changes in animal health can be monitored in a novel fashion to significantly prevent health and economic loss.

2.3. Detecting subclinical ketosis using microfluidic biosensors

Regular monitoring of clinically important β -hydroxybutyrate BHBA to provide early diagnosis of Subclinical Ketosis (SCK) is essential for management of dairy cattle health. Early detection of SCK helps reduce the risk of the disease progressing into a clinical stage. Previously the literature focused on the diagnosis of diabetic ketoacidosis, specifically in humans. This method is inaccurate because it fails to account for the

multiple blood groupings in cows. Although both cows and humans are mammals, they have considerably differences in their basic physiology. There are 11 major blood group systems in a cow, whereas there are only four blood group systems in humans. Due to variation in the blood group system, there is a contrast in the antigen expressions in cows and humans which makes it unsuitable to use human ketosis detectors in cows for the determination of β -HBA. Therefore, the reliable measurement of β -HBA in animal samples is possible only through highly specific and sensitive sensors which have been built based on microfluidic systems [45]. The early and efficient detection of SCK can be achieved by using these robust and movable devices. This will not only help in the prevention and control of ketosis, but significantly contribute to the health management of dairy animals [46].

Microfluidic technology is an effective way for on-farm detection of this disease. Recent prototypes seem promising, and are cost effective as compared to the conventional laboratory methods like chemistry analyzers and microplate readers. In one study, real-time determination of β -HBA was set as a prime indicator in the diagnosis of ketosis, and the miniaturized biosensor was characterized by high sensitivity and specificity towards β -HBA, with a detection limit of 0.05 mM. The developed biosensor used the spectroscopic principle of the absorbance of UV in 445–455 nm range. The β -HBA concentration in samples is depicted by the intensity of light signals transmitted by the Si photodiode. Detailed analysis of the light absorption was performed by a custom-built optical biosensor [46,47]. Veerapandian et al. described an electrochemical biosensor platform that can selectively detect β -hydroxybutyrate by immobilizing the enzyme 3-hydroxybutyrate dehydrogenase. Xuan Weng et al. also developed a rapid, low-cost microfluidic biosensor with high sensitivity and specificity. The microfluidic biosensor showed a response time of 1 min and a detection limit of 0.05 mM concentration [47].

2.4. Farm monitoring

Traditional farm monitoring, such as using written notes or a simple device without data sharing capabilities, is an inaccurate method with high probability of human error. Previously, the use of Global Positioning Systems was proposed, but it required detailed field maps and was costly due to the involvement of transmission of data from satellites. Voice entry systems also has its drawbacks because of noisy backgrounds in farms and fields. A recognition method for farming operations using Radio-frequency identification (RFID) has also been proposed. RFID tags attached to farm animals can record simple farming tasks. RFID tags can also be attached to or embedded in animal bodies, tracking such health control factors as fattening management, milking management, and behavior [24]. Tagging animals has now become a trend, as millions of fish, bees and even racing pigeons have been tagged to keep tabs on their locations [2].

RFID tagging, which is being employed in different diagnostic devices including implants and collars, has recently merited attention in the international animal market. There has, however, been a legal concern related to the manufacture and sale of these advanced products, since some dog training collars have been used to deliver electric shocks to dogs [26]. RFID-based systems require fixed, coordinated pressure sensors. Banhazi and Black [30] have discussed the need to integrate data measurement and acquisition systems, as well as protocols to identify inefficiencies and automated decision making.

The principles of Total Quality Management (TQM) and Hazard Analysis Critical Control Point (HACCP) form the basis of Precision Livestock Farming [30]. Precision Livestock Farming (PLF) has revolutionized the livestock industry and contributed towards animal welfare. It is a system which comes with many benefits and ensures maximum use of all resources, thus controlling the health status of animals. PLF helps bring maximum productivity, even in a varying environment, through the sensible use of feed and water for animals (Table 2). It helps utilize animals for the benefit of humankind through production

of high quality results. PLF makes the real-time supply chain management throughout the animal industry possible. This system integrates electronic technology in farming and ensures that the information obtained through measurements is used in a way benefitting farmers. PLF has helped reduce greenhouse gas emission, enabled better marketing of livestock products, and enhanced the economic development of rural areas. Reliable statistics from farms and optimization of the livestock feed, depending on the feedback of different farms, can lead to more successful farms and help make smart business decisions [48–50].

PLF works for animal welfare through a variety of devices, including automated tools that integrate audio- and video-captured data for early disease detection and warning systems (Fig. 3). The sound and image data is analyzed by animal experts into a database used for creating suitable algorithms [51,52].

A technology called Flockman™ is a recent and innovative feed control system for broiler chickens. This PLF system monitors the regular feed intake of living birds and allows the farmers to alter the feeding system as needed. A system that determines the growth trajectory of the body mass of chickens and controls the feed intake is also being used. This technology has significantly reduced the mortality rate of broiler chickens. Flockman also analyzes the impact of different environmental factors in the facility, such as heat, ventilation and humidity, on the growth and health of chickens [53].

Subcutaneous biophotonics sensors can help track the hemodynamic parameters of animals. It uses photodetectors and surface-mount light sources, like glass capsules used for microchip implants. These sensors use reflectance-based pulse oximetry measurements to track cows' pulses [54,55].

In contrast to previous approaches, PLF systems allow real-time monitoring of animals so immediate action can be taken by farmers in support of animal health and better product yield. A recently developed system called the eYeNamic system automatically monitors the behavior of housed chickens. The spatial distribution of birds allows the calculation of zone occupation index and zone activity index of the broilers with the help of top-view cameras. PLF technology clearly has great potential as a livestock management tool for farmers [6,29,56].

Sensor systems for measuring fat and protein content in milk are used frequently on farms nowadays. The sensor system used differs

according to the milking systems used on each farm. These sensors provide health and fertility data of cattle. Reproductive performance in dairy herds can be analyzed through estrus detection. Sensor systems have been reported to detect roughly 80–85% of cows in estrus. It is not fully determined whether the use of sensor systems also benefits health and production of cows. In older studies, it has been proved that higher estrus detection resulted in a shorter calving interval, consequently leading to increased milk production. High somatic cell count has been linked with lower milk production. Usage of automatic milking systems has proved to increase milk production. Different statistical analyses can be used to see the role of sensors for mastitis and estrus detection for dairy cows. Fat, protein, temperature and milk temperature sensors can also be used [57,58,59] to enhance the animal production systems. Studies have suggested that dogs can be trained, through positive reinforcement and an optimized training protocol, to sense the differentiation of vaginal mucus samples from cows which are in estrus and samples from cows in diestrus [60].

Better reproduction rates in cattle can be obtained if hormone levels are measured efficiently. Using Electrochemical Impedance Spectroscopy (EIS) techniques, which can detect progesterone in purified water, is one solution. Planar capacitive sensors use silicon substrate, thin-film microelectromechanical-based semiconductor device fabrication

Table 2
Recent developments in PLF systems.

PLF technologies over the years	References
Weight estimation of pigs via vision tools	[9]
Cattle monitoring system and tracking dairy cow behavior	[96,66,64]
Cough analysis in animals using audio and video data for identification of respiratory infections	[11,12]
Sound analysis in cows	[14]
Detection of pig screams	[91]
Stress detection in laying hens	[10]
Automatic detection of cow's oestrus in audio surveillance system	[90]
Noise analysis to evaluate chick thermal comfort	[93]
Sensor and instrumentation for progesterone detection	[61]
Wireless system for pregnancy detection in cows by monitoring temperature changes in body	[92]

Table 1
Analytes and methodologies involved in sweat analyzers.

Bio fluid	Detection method/Platform	Analytes	References
Sweat	Conductometric sensor based on poly-(2-acrylamido-2 methylpropane sulfonate) connected to impedance meter	pH, Cl [−] and Na ⁺ concentration (help in clinical management of Cystic Fibrosis)	[34]
Sweat	Multi-device characterized by three fringing field sensors	Glucose (role in diabetes management)	[34]
Sweat	Device based on an amperometric biosensor characterized by a graphite electrode with embedded alcohol oxidase, horseradish peroxidase and ferrocene. It is connected to a miniaturized potentiostat and a microprocessor	Ethanol	[34]
Sweat	Electro generated chemiluminescence (ECL) biosensor using a luminol hydrogen peroxide sensitive compound, lactate dehydrogenase and pyruvate oxidase (as catalyst), adsorbed onto a carbon nanotubes layer.	Lactate (indicates the switch from aerobic to anaerobic metabolic conditions in sports science applications)	[34]
Sweat	Textile-based fluid handling system made of a moisture wicking material (mixture of polyester (92%) and lycra (8%)), and an optical pH sensor based on bromocresol purple with the optical detection system	pH	[34]
Sweat	Potentiometric sensors	Ammonium, pH and Na ⁺	[34]
Sweat	Amperometric sensor	Lactate	[34]
Sweat	Fully integrated mechanically flexible sensor array for multiplexed in situ perspiration analysis	Sweat metabolites (such as glucose and lactate) and electrolytes (such as sodium and potassium ions), as well as the skin temperature (to calibrate the response of the sensors)	[36]
Sweat	Wearable electrochemical sensor e.g. temporary tattoo-based printable stripping-voltametric sensor	Trace metals like zinc	[44]
Sweat	Microfluidic models.	Ions (Na ⁺ , Cl [−] , K ⁺ , NH ₄ ⁺), small molecules (ethanol, cortisol, urea, and lactate), and peptides (neuropeptides and cytokines)	[37]
Sweat	Standard laboratory procedures for calcium detection or calcium level tests for cows like a Water hardness test kit or a suitable test strip. Other tests like titrimetric, ion selective electrodes and photometric determination methods can be used as well.	Calcium	[38]

technology. This sensor can evaluate conductivity, permeability and dielectric properties of the reproductive hormone progesterone and its concentration quantification in purified water [61]. Integrating wireless sensors for online health monitoring systems has been designed and been investigated as monitoring systems [62–66]. Proportional Integral Derivative (PID) control technique has also been applied to reduce labor and yield higher profits in poultry feeding management [67].

2.5. Pathogen detection

2.5.1. Detecting influenza virus using FRET

Influenza virus is a highly contagious disease among birds. It can spread through saliva, nasal secretions and other excretions from infected birds [69].

A homogenized and uniform fluorescence-quenching-based assay has recently been developed which specifically detects influenza virus surface antigen hemagglutinins (HAs). This assay will revolutionize the process of detecting viruses, and can be a sensitive diagnostic tool for influenza virus discrimination. The assay consists of two nanoprobes: the glycan-conjugated highly luminescent quantum dots (Gly-QDs), and the HA-specific antibody-modified gold nanoparticle (Ab-Au NPs). These nanoprobes are brought together when exposed to strain-specific HA. This happens due to a very specific binding event between the HA and the two nanoprobes, which forms a sandwich complex. QDs fluorescence intensity is hence diminished, as a non-radiative energy transfer occurs from QDs to Au NPs [70]. The fluorescence changes, and a resulting correlation between the targets HA concentrations are easily observed. Moreover, HAs origin can be detected easily and, due to the specific interaction between HA and glycan with sialic acid residues, we can distinguish between the human (H1) and avian (H5) viral subtypes. Normally, the influenza virus is detected by the so-called “gold standard” viral culture and RT-PCR, which is time- and labor-intensive. The immunoassays like ELISA are less specific and less sensitive, which makes them inconvenient to use for this purpose. However, fluorescence resonance energy transfer (FRET) is sensitive enough to detect small changes in interaction between the biomolecules. It is a very powerful method in which non-radiative energy is transferred from an excited donor to an acceptor at ground state (quencher) placed almost 1 to 10 nm away [70]. Illegal feed additives can also be detected using fluorescence quenching and immunochromatography; e.g., determination of ractopamine and clenbuterol in porcine muscle and swine urine [71,72].

Direct contact with infected poultry can infect healthy birds with avian influenza virus. Even sharing water and feed can result in contamination. The disease manifests itself in two forms, with low and high extremes of virulence. The low pathogenic form is less virulent and shows mild symptoms, like lesser egg production. The highly pathogenic form manifests itself by rapidly spreading through flocks and affecting multiple internal organs. A system called Sensor Networks assists in collecting data from a desired location and transmitting it to a computer. Sensor Networks contains different sensors which perform extraordinary signal, alarm, and decision-making functions. This sensor system uses open source technology—Arduino—an electronics prototype that provides parameters like body temperature and movement of the animals. The alarm is sent to an observing station, which is used to monitor whether the observed value is lesser or greater than the normal value. The detection of avian influenza is thus assisted by monitoring the level of H5N1 virus, which causes mortality in chickens. The use of easily implementable ZIGBEE wireless protocols also helps in constant monitoring. Wireless Sensor Networks can be enhanced through antennas and amplifications for long-distance applications. The ADXL320 accelerometer detects chicken motion, feeds it to Arduino, and records the readings [8,68,73–75].

Other viruses linked to reproductive or respiratory diseases, such as Porcine Reproductive and Respiratory Syndrome Virus (PRRSV), can also be detected using biosensor-based Imaging Ellipsometry [76].

2.5.2. Detection of bacteria using SERS

SERS, a label-free biosensing method for bacterial detection, provides information about the chemical structures of analytes. SERS uses the intrinsic vibrational fingerprint of analytes to detect molecules. Moreover, the performance of Raman spectroscopy remains unaffected by the surrounding water, allowing easy bacteria detection [21]. Raman spectroscopy has found its application in even the label-free modes in the analysis of chemical and biological components. The detection of disease-causing bacteria in drinking water, using the label-free near-infrared surface-enhanced Raman scattering/spectroscopy (NIR-SERS) method is a recently proposed technology, and provides a diagnostic platform. This analytical method is a rapid way for the successful label-free identification of pathogenic bacterium in health-care applications. The in situ synthesis of silver nanoparticles (Ag NPs) within the bacterial cell suspensions provides a means for the detection of foodborne bacteria. This method requires no preparatory phase and is label free. To enhance the assay's sensitivity, Triton X-100 is used to pre-treat the bacterial cells. However, probing the fingerprints of bacteria through this method becomes difficult due to poor selectivity, as it is a simple mixing process. This remarkable technology not only helps detect the pathogenic bacteria, but also enables the discrimination between different types of these bacteria, such as Methicillin-resistant, *Pseudomonas aeruginosa*, *Escherichia coli*, *Staphylococcus aureus* (MRSA), and *Listeria* spp. This method also helps to differentiate between two species—*L. monocytogenes* and *L. innocua*—through the comparison of the SERS spectra and Raman frequencies. Similarly, it is also possible to discriminate between two MRSA strains from clinical isolates [77]. Even the most intricate details, like the molecular composition of a sample, can be revealed using this spectroscopy technique at a micrometer scale. Metallic nanoparticles (NPs) on the order of 104–106 nm were used with SERS via SPR. This method makes sure there is homogenous contact of constituents of the bacterial cells to nanoparticles, and provides an intense spectrum with better selectivity [77].

Another study explored a novel microfluidic platform that employs methodologies for chemo metric data analysis, including a combination of principle component analysis and linear discriminant analysis, as well as silver nanoparticles. Distinguishing eight key foodborne pathogens (*E. coli*, *L. monocytogenes*, *L. innocua*, *S. typhimurium*, *S. enteritis*, *Pseudomonas aeruginosa*, MRSA 35 and MRSA 86) which significantly affect the food industry has been made possible through this method. Surface-enhanced Raman spectroscopy technique has made the imaging and detection of gram-negative and gram-positive bacteria successful as well. The discrimination between two types of bacteria depends on the difference in the scattering intensity of gram positive bacteria, which is higher than that of the gram negative bacteria. Another method for bacteria detection suggests the synthesis of magnetic-plasmonic Fe₃O₄-Au core-shell nanoparticles to concentrate bacterial cells. This is accomplished through the application of an external point magnetic field and SERS [21].

Biosensors can also be used to detect live bacteria in drinking water using Ag nanoparticles. This novel technology has also detected anthrax spores on nanosphere substrates. The-multi drug-resistant strains of bacteria can also be detected using complex nanohybrid systems. These systems are developed by combining antibody-conjugated gold nanoparticles with single-walled carbon nanotubes.

2.5.3. Detection of pathogens using HNPs-GO electrodes

HNPs-GO electrodes, known for their remarkable electrochemical immune-sensing properties, are being employed to detect *Listeria monocytogenes* (Lm), a major foodborne pathogen. Lm is a gram-positive bacterium that causes listeriosis, which is very prevalent and has a high mortality rate. This alarming situation calls for an efficient system to detect Lm in food products. Nanosheets of graphene oxide (GO) coated with the hybrid nanoparticles of a silver-ruthenium bipyridine complex (Ag@[Ru (bpy) 3]²⁺) core and chitosan shell have significant immunosensing properties. The oxygenated groups of GO and the

amine groups on the surface of the hybrid nanoparticles play a significant role in this unique application. Contamination in milk and other food products can be easily detected using monoclonal antibodies and HNP-GO immunosensors. Hence, intelligent and specific optimization of the bio-recognition elements on the HNP-GO electrode has a bright future in the food processing industries. Having good fabrication, thin layering and tunable oxygen functional groups make two-dimensional graphene oxide (GO) and reduced GO (rGO) preferable. Surface treatment of the active components on GO nano sheets change the crystallite size and related properties. Strategies like elemental doping and photoirradiation are applied to vary the physico-chemical functionalities of GO. Biosensing applications can be improved to a great extent by incorporating a durable single hybrid nanostructure on the GO surface with suitable optical and biocompatible capabilities. Reactivity of the oxygen functional groups present on the edges of GO govern the chemical functionalization of the materials on GO's surface [78]. The detection of pathogens from the skins, oral cavity, feces of farm animals and also in the environment of the barn or pen can be efficiently quantified using the hybrid nanoparticle based biosensors.

2.5.4. Detection of infectious agents

Globalization has led to the rapid and unhampered distribution of animal products all over the world, posing great threats to humans. Transboundary animal diseases (TADs), including foot-and-mouth disease and classical swine fever, can spread very quickly across borders and countries. These diseases affect the animal trade and have a devastating impact on animal husbandry. Some animal diseases cross the species barrier and can affect humans, causing zoonotic infections. Therefore, appropriate methods must be applied for the diagnosis of such diseases. This will help in devising special precautionary measures like vaccinations and quarantine.

In direct detection methods, infectious agents can easily be detected in samples collected from animals. Classical methods for detection of microbes include identification by culture techniques and immunofluorescence. Molecular techniques include Polymerase Chain Reaction (PCR) and loop-mediated isothermal amplification (LAMP). Occurrence of infections in hosts can be diagnosed through indirect methods as well; for example, by identifying the antibodies against various infectious agents.

The advantage of PCR is its high specificity. PCR is a highly sensitive assay that can diagnose infectious agents at molecular levels. Since every microorganism has its own unique genome, PCR enables amplification of the genetic material, including DNA and RNA. The real-time PCR technique has many variants, e.g. FRET-based assays, TaqMan assays, etc. SYBR Green is a cost-effective method, since it avoids the use of probes. PCR techniques are thus affordable, and are put to use in lightweight portable devices for on-site infection detection. Use of novel isothermal amplification methods further facilitates the on-site diagnosis of infections in animals. For example, loop-mediated isothermal amplification (LAMP) runs at a single temperature level and gives results readable by the naked eye. Simultaneous detection of different infectious agents is also made possible using various technical approaches, such as the padlock probes and various liquid microarray readouts of PCR results [79].

Enzymes used in PCR include polymerases, reverse transcriptases, nucleases, etc. Use of thermostable polymerases allows amplification through thermocycling. The PCR product can be visualized through agarose gel electrophoresis using fluorescent dyes. Electrophoresis allows estimation of the amplicon length and maintains the specificity. But using gel-based PCR can sometimes be laborious, and doesn't allow the quantification of the initial viral load. Due to lack of specificity, problems like false positive detection arise. This problem has been solved with real-time PCR, which allows a closed-tube assay with minimum risk of cross contamination. In this assay, the product is monitored during reaction by DNA-binding moieties that bind to the amplified DNA and emit fluorescence without the requirement of the step of gel

formation. The cycle number at which the fluorescence reaches its threshold level depends on the initial viral load. Communication of laboratory results with the health authorities in a rapid manner can lead to the successful eradication of infectious diseases from animals; rapid two-way communication between laboratories and the practitioners will ensure the success of control program. Three final PCR technologies aiding the detection of infectious agents include MAP, the Field Effective sensor, and Vantix.

John's disease (JD) is a major gastrointestinal disease of cattle caused by *Mycobacterium Avium* subspecies *Paratuberculosis* (MAP). This cattle disease causes premature culling and reduced milk production. This disease can be controlled using conductometric biosensors which combine immunomigration technology with electronic signal detection [80].

The Field Effective sensor can be used for direct serological analysis. BHV-1, the pathogen causing bovine respiratory disease, can be detected through this method, which is faster than ELISA and plays an effective role in disease intervention [81].

Bovine herpes virus-1 antibodies can be detected by the biosensor assay, which provides quantitative analysis of antibodies in milk samples. One such biosensor assay, Vantix™, has been developed recently, and provides a platform for rapid routine immunological testing [82].

2.5.5. Hybrid technology

There are fair chances that bacterial concentration can obscure the relative peak and baseline intensity of the spectral data from the biosensor. A hybrid technology has been proposed to avoid this. The uniform concentration of bacteria can be achieved through the modification of substrate surfaces, using microfluidics to avoid spectral interference in RAMAN spectra from mixed bacterial samples. Miniaturization of the RAMAN spectrometer is needed to further develop this technology. The integration of the RAMAN spectrometer and the microfluidic device platform faces various challenges, but must be considered for the successful application of NIR-SERS detection of foodborne pathogens [77].

Another method, using electrokinetics-enhanced capacitive immunosensors for point-of-care serodiagnosis of infectious diseases, has been reported. Capacitive bio-affinity detection using microelectrodes is considered as a promising label-free method for point-of-care diagnosis, though it has challenges in sensitivity and time [83].

2.6. Movement and behavior

Movement and behavior of farm animals can relay information about their level of activity and well-being. Moreover, physical defects in limbs can be detected early by abnormal movement. In large farms, relying on naked-eye observation can result in human error and a delay in diagnosis. Therefore, better methods for observing farms should be incorporated. For instance, a top-view camera may be used to analyze motion and detect low weight in pigs. The automatic detection of a low-weight pig through this method does not require any manual inspection by a farm administrator (Fig. 5) and hence decreases labor for farmers. By using motion detection technology and recorded video, object areas are identified using Hue saturation values (HSV) color information. This detection technique can also identify a group of touching pigs standing close together. Gaussian Mixture Model (GMM) is applied for the detection of and determining the size of individual moving pigs. The partially and fully moving pigs can also be identified using this model and hence, low-weight pigs can be identified without human intervention [9]. GPS and 3D collar-mounted accelerometers have also been developed [84,85,86].

A technology named MooMonitor is also helping farmers determine the health of their cows through measurement of the physiological conditions of individual farm animal. This technology makes use of wireless sensors that allows the farmers to detect individual cow heats and health events with ease through data analysis. Heat in cows and their illnesses can thus be recognized through this device which can help the

farmers keep their livestock ideal and healthy. A similar technology, Silent Herdsman, has been specially formulated for farmers to be used on farm animals. Like a collar, it's wrapped around the neck of each animal and supervises all activities of cows and their behaviors. First it records behavioral patterns, and then it detects and records any changes in those patterns. In this way, estrus cycles and onset of certain sicknesses can be successfully identified and monitored.

The Royal Botanic Gardens in Kew, United Kingdom uses micro-technology to track the movement and locomotion of honey bees and their pollination activities. It is believed that this can provide breakthrough research on the constantly declining population of honey bees, which are a valuable asset for humans. Australia's Commonwealth Scientific and Industrial Research Organisation (CSIRO) is also interested in solving the rising issue of a sharp and continuous decline in honey bee populations. A large number of Australian honey bees have been tagged with microsensors to analyze the lifestyle and behavior of hives. This technology, Swarm Sensing, is assisting Australian entomologists determine the patterns of honeybee movement [87].

An Israeli company is currently offering a full array of GPS systems to make the detection of predators easy. It also sends drones from phone to record the video of animal grazing and forms a fence around animals so they don't wander off [28]. Motion History Image-based technology (MHI) is an innovative and reliable method that isolates the shaking motion of a coughing from other motions [11]. Image-based video analysis can help farmers isolate a coughing animal from its herd.

Computer vision technology is based on the real-time monitoring of pigs based on the circadian rhythms. Camera sensors can easily detect the relationship between the dust concentration present in pig barns and pig activities. Different image-processing systems can be employed to classify the thermal comfort states of pigs and analyze their behaviors. The circadian rhythms in pig barns without windows, and with 24-h light-on conditions, can be investigated. This system of visual streaming data can analyze the activities of weaning and individual pigs as well [88,89].

2.7. Stress detection

Stress has deleterious effects on the productivity of commercial chickens, so a quick, accurate stress measurement technology would be advantageous. Sound analysis is a reliable measure to indicate stress in chickens. An inexpensive and automatic prototype has hence been developed recently to notify farmers to stress levels using sound data. Its structure consists of three binary-classifier support vector machines. The sound emitted by the hens is first detected; then the classification module identifies and classifies the stress in the sound. An experimental evaluation is then prepared, using real-time sound data from an audio surveillance system. This model has been validated at 96.2% accuracy and is a very precise classification model [10].

Lactate is an important indicator of stress in animals, as it plays a significant role in tissue homeostasis. Being a product of anaerobic and aerobic glucose metabolism, it is a main component of glycolysis. During stress, excess pyruvate is produced during glycolysis, which is converted to lactate and stored in tissues or exported to blood. This increased level of lactate in blood and tissues can be detected and used as an indicator of stress. Amperometric biosensors have gained popularity, since they are used for lactate sensing. These biosensors are composed of electrode material which is chemically modified and has biological recognition elements (BRE). Moreover, body-sensing technology senses various metabolites and electrolytes using Band-Aid like RFID sensor patches and temporary tattoo-based sensors.

The need for power, short battery life, and packaging material are major disadvantages of conventional methods. Now, using mobile phone and smartphone apps, the data can be transmitted wirelessly. However, wireless sensors also need batteries, and the replacement of these batteries becomes a time-consuming task. These batteries are also environmentally unfriendly and need to be recycled. Therefore,

there is a need for self-powered biosensors. Using battery-free sensors fueled by a biofuel cell is an economic approach, since these biofuels use enzymes. The discussion of these enzyme-based biofuels in sensors gained momentum more than a decade ago. Lactate levels can be analyzed in sweat using an enzymatic sensor. This sensor is NAD⁺-dependent and has a biofuel cell as its power source. This sensor patch has reportedly excellent performance, since the lactate dehydrogenase-based sensor component shows linear current response with increasing lactate concentrations. The sensor, which was coupled with the power source, energy harvester and micropotentiostat, is a great success. Connecting the sensor with the Micro Potentiostat makes the chronoamperometric detection of lactate relatively easier and possible. This Sensor Patch System has been tested under laboratory conditions and its performance has been verified [4].

2.8. Sound analyzers

The sound an animal makes can give a hint to its physical well-being. Some sounds are used to attract mating partners and some to threaten predators, while others can indicate respiratory diseases [13,16,90]. For instance, a rumination microphone helps to observe cud chewing in cows [28]. Technologies like deploying cameras on pets or farm grazing animals and analyzing their sounds (like barking and or coughing) can be used efficiently to keep check on the animals so they don't go missing or wander off [1].

Sound analyzers can also be used to monitor pigs' health and body conditions. Their sounds help farmers evaluate whether the animal is under stress or not. The interference of environmental and background noise can hinder the detection of pig screams. An automatic scream-detection method has been devised recently which distinguishes the screams of pigs from other sounds on the basis of sound structure, power, frequency, duration and variability. By using different algorithms and differentiating between the spectrograms, this classifier is a big breakthrough in animal health management [91]. Respiratory diseases can also be detected using sound [12].

The sound data from a group of Korean native cows (*Bos Taurus coreanae*) has helped in detecting the abnormalities in the oestrus cycle of cows [90]. An automated analysis of these anomalies using a sound sensor has contributed significantly to livestock management. Using remote sensing techniques, vocalizations can be used to recognize the weaning and hungry states of cows as well. Previously, research has been done to extract meaning from specific calls of cows and accurately evaluate the emotional state of a cow.

Bird vocalizations can be used to estimate thermal comfort for chicks during the heating phase (Fig. 4). Group behavioral patterns and their vocalizations were found to be correlated not only by the noise amplitude, but also by the noise frequency spectrum. Such noise sensors may be important to determine the effect of environment on chick health [93]. Analyzing and correlating the sound frequencies of different vocalizations during the life of birds can serve as a very useful tool for the detection of growth of chickens [11,94,15].

Wasting disease is a potentially risky and highly contagious condition in livestock which requires 24-h monitoring for timely detection of disease. This disease can be detected successfully by obtaining and using audio and video data. The cough sound can be detected through audio analysis and motion detection [95].

2.9. Determining metabolic activity

Metabolic disorders in cows can be detected by increased concentrations of non-esterified fatty acids (NEFA) in biological fluids, which act as significant biomarkers. Cost-effective and bio-friendly sensor elements based on ruthenium bipyridyl complex-modified graphene oxide nano sheets ([Ru (bpy) 3]2p-GO) can be used for early diagnosis of metabolic diseases of cows. These biosensors can be used for Circulating Non-Esterified Fatty Acid (NEFA) detection through electrochemical

analysis. [Ru (bpy) 3] 2^{+} -GO electrodes have reliable redox properties as compared to GO electrodes. NEFA levels are a good indicator of negative energy balance (NEB). During NEB, adipose fat is mobilized as NEFA and transported to the liver to be oxidized or re-esterified into triglycerides. In the liver, excessive NEFA might increase the risk for clinical malignancies such as fatty liver, ketosis, displaced abomasum, metritis, and retained placenta. NEFA has a prime economic value for livestock producers and also indicates fertility issues in cows. NEFA represents a dairy cow's health and plays a crucial role in the health management of livestock.

Carbon electrodes are the electrochemical-sensing platforms and have the ability to be used in the robotic milking machine and other important diagnostic systems. Redox active hybrid GO materials, being cost effective and electrochemically stable, are actively used in the bio detection applications. Newly-fabricated nano-biosensors have iron-containing enzymes like lipoxygenases, which form acid peroxides by catalyzing the oxidation of polyunsaturated fatty acids. Lipoxygenases have a future in scalable bio-catalysis. Using the GO nano sheets allows electrochemical detection of NEFA; the lipoxygenase immobilized on the electrode surface catalyzes the NEFA into fatty enones and affects the redox reaction occurring in interface. This electrode system is linearly dependent on different concentrations of the standard NEFA, as well as serum samples, hence providing accurate data for detection of metabolic disorders of cows [96]. Increased milk production and body temperature in cows after insemination acts as an indicator of the mother's immune response to the embryo's entry into the uterus. Hence, low-cost electronic components and materials could be used as non-invasive pregnancy detection in cows without human interference [92,61].

2.10. Detection of toxins

Microfluidic devices allow effective toxin and antigen detection, which has many beneficial applications in health care for animal health. Neurotoxins can be detected using microfluidic devices. Recently, the National Center for Food Protection and Defense in St. Paul carried out extensive research and developed a highly sensitive microfluidic platform for detecting botulinum neurotoxin in solution. These microfluidic devices are composed of input and detection ports connected by a microchannel.

Microfluidics are also being used in the food and dairy industry, where liquids and solids are blended together to manufacture dairy products. Microfluidics technology is very helpful in the formation of homogenous liquid mixtures, keeping viscosity and density balanced. The valuable microfluidics technology can also be combined with food processing equipment to make suspensions of various compositions. Microporous calcium alginate gels, low-energy food products, foams and emulsions can be produced through this technology as well. Nanoparticle synthesis and formation of plastic microfluidic chips for applications in food safety has also increased recently.

Microfluidic devices are being employed for culture and manipulation of embryos in assisted reproduction of cattle [97].

Quantitative analysis of the proteins wheat gluten and Ara h 1 can be done by the use of a microfluidic ELISA platform combined with a recently developed custom-designed optical sensor. This microfluidic ELISA biosensor dramatically reduces the total assay time from hours to minutes and decreases the consumption of sample/reagents compared to commercial ELISA. Therefore, this technology has a big edge over commercially available conventional methods like dipstick tests. It is hoped that this detection technique will soon be commercialized and applied in food control agencies for better human and animal health. Technologies which only use microfluidic chips require expensive and heavy instruments, and thus cannot be used as portable devices. Integrating microfluidics and biosensors has helped in the development of point-of-care (POC) diagnostics. This useful ELISA

device can be used by food safety inspectors and industries in favor of better health management [98].

L-glutamate is an excitatory neurotransmitter, and its regulation is related to important cognition processes. Different electrochemical sensors can be implanted in the bodies of animals for its detection. Microsensors are used for this purpose, but these sensors pose difficulties, as they are susceptible to interference by other chemicals, and yield small signal responses. Current commercial recording systems are inefficient, as they restrict the movement of subjects. Hence there is a need for a wireless system which reduces interference and enables data transmission to the Internet or smartphone [99].

2.11. Detection of temperature

Body temperature of animals is a very important indicator of their physiological well-being. For this purpose, thermistors, thermocouples and infrared radiation sensors can be used. Coupling these technologies with user-friendly interfaces can lead to development of new portable gadgets for farmers, contributing to PLF [7,100]. Data obtained by studying the thermoregulation process in animals with factors influencing the body temperature, like pregnancy, parturition and lactation can be used to get an idea of basic status of animal. Body core and basal rectal temperatures can be used to analyze body temperature variations and link them with various abnormalities. Body core temperature reveals the temperature near major organs of body, such as the heart, viscera and brain. This temperature is measured by rectal, vaginal, vascular and digestive-tract sensors. Mid-peripheral temperature, on the other hand, is the temperature of the body parts intermediate between body core and surface sites. It is thus evaluated through intramuscular chips. Peripheral temperature is measured through an animal's outer surface; microchips are embedded a few centimeters deep into skin to measure this temperature. Similarly, infrared thermal imaging cameras serve as infrared radiation thermometers which measure body temperature at various points, thus producing a two-dimensional image called a thermogram. This software-intensive technology allows monitoring the body temperature of animals in different environments [5].

Recently, *E*-Pills have been used to track health data. These pills stay in a cow's rumen for a very long time and transmit data through cloud software. In this way, the farmer can successfully collect the important data regarding body temperature, heart rate, etc. of the animal [1]. Similar sensors have been developed for non-lactating dairy goats [101]. The use of e-tags, which monitor body temperature to detect disease onset, is also becoming common.

2.12. Saliva analyzer

Biological fluids of living beings, like tears, sweat, and saliva, can be used for testing health and detecting pathological conditions. Similarly, breath and interstitial fluids of the body can also be used for this purpose. Noninvasive monitoring of uric acid in saliva can be done using a mouth guard with an integrated screen-printed electrode system. The uricase enzyme is utilized in this system, which uses electronics (potentiostat, Bluetooth and microcontroller). Usually, biosensors require a lot of power. However, this platform is capable of transmitting information to laptops and smartphones, where the information can be processed and stored. This mouth guard biosensor is highly selective and stable for uric acid detection in saliva, since it covers a large range of concentrations. This real-time biosensor is a wearable monitor being employed in different health applications [35]. Analyzing saliva is a non-invasive, readily available method. It is extremely useful in analyzing mouth conditions and Gastrophageal Reflux Diseases (Table 3).

Monitoring lactate variations in saliva is another practice used to detect health conditions in animals. Materials like carbon nanotubes and graphene can be employed in the production and promotion of such health-care related technologies. Wearable "lab on a chip" systems are gaining popularity among animal handlers. Combining textiles and

Table 3
Selected wearable salivary electrochemical sensors.

Bio fluid	Methodology/Platform	Application	References
Saliva	Enzyme (uricase) modified screen printed electrode system integrated onto a mouth guard platform along with anatomically-miniaturized instrumentation electronics featuring a potentiostat, microcontroller, and a Bluetooth Low Energy (BLE) transceiver	Uric acid detection in saliva for various health applications.	[34]
Saliva	Microfluidic health monitoring device in a lollipop (Lollylab system) using saliva as sample. The chip is embedded with a candy shell that includes saliva stimulants.	Disease monitoring, pregnancy testing, hormone monitoring, detection of virus and strep throat infection in livestock animals, and monitoring of medications	[98]
Saliva	Carbon nanotubes and graphene	Analyzing the mouth conditions and Gastrophageal Reflux Diseases. Monitoring lactate variations in saliva is another practice which is being used for the detection of health conditions in animals	[39]
Saliva	Partial chrome-cobalt denture employing potentiometry	Fluoride detection using Lanthanum fluoride	[34]

sensors lead to more robust, mechanically strong microfluidic devices. Polymers like Kapton and Mylar are used because of their impressive thermostable characteristics. Other materials, like GoreTex, combine hydrophobic properties with water permeability. Similarly, chemical and biosensors can be brought together, as in the case of Google Glass, which can collect all the sensor data from a body. Such novel wearable technologies are undoubtedly opening avenues for facilitated animal health management. Glucose can be successfully detected in interstitial body fluids and is an analyte of great interest. Other analytes, like ethanol, can also be quantified through skin poration. This technique does not cause pain and acts as an interstitial harvesting system.

Breath monitoring for clinical analysis and therapy is catching the interest of livestock handlers as well. The amounts of oxygen and nitric oxide in breath give an understanding of health conditions. The vapors emitted from animal's breath makes the noninvasive detection of different analytes possible. However, there are challenges associated with this method as well. These include humidity interference and insufficient accuracy of breath measurement methods. Portable hand-held breath analyzers are quite helpful in this regard. These sensors are based on basic electrochemical techniques. They contain a pressure meter, spectrometer, and a Teflon piece attached to a Mylar balloon for breath sample collection. Many monitoring tools, like oxygen breath analyzers, contain sensors based on Organically Modified Silicate, or sol-gel. Ruthenium oxygen-sensitive luminophores and fluorophores produce fluorescence emissions after interacting with oxygen. These devices have an appreciably good response time which make them preferable.

Ammonia can also be detected in breath, and is an indicator of many stomach infections. Ammonia can also give insight into the respiratory system of animal, since it diffuses out of blood and travels to lungs. Similarly, pH can also be monitored with the help of sensors. Such sensors use textile-based fluid-handling systems along with optical pH sensors containing bromocresol purple. The sensors measuring sodium levels in sweat contain Ion Selective Electrodes. Some wearable technologies (Table 4) also contain attached Bluetooth devices. The introduction of screen-printed technologies that attach electrodes to skin are also gaining success. Potentiometric sensors for the measurement of ammonium and amperometric sensors for lactate measurement have also been reported [39].

2.13. Monitoring of metabolites

Monitoring metabolic activities of animals is important. Integrating bio-nanosensors to detect metabolites like lactate, glucose and ATP is being evaluated with animal models. These sensors contain electrochemical components combined with a radio frequency communication system with an antenna. This system has proved to be reliable when tested on animal models. These bio-nanosensors have also been tested for inflammation in tissues when introduced in the animal models. No effect was seen on the concentration of ATP in the subcutaneous micro-environment by suture. However, neutrophils infiltration is detected by invasive procedures, like puncturing for biochip or nano-biosensor implantation of Nano-Bio-Sensors [102]. After hemorrhage, the intestinal tract is the organs which is involved in an ischemic injury, which can

Table 4
Disease detection through breath analysis using various methodologies.

Disease/Infection	Methodology	Biomarkers	Type of sample	References
<i>Mycobacterium tuberculosis</i>	Enose	VOCs pattern recognition	Breath	[108,117]
<i>Pseudomonas aeruginosa</i>	Enose	VOCs pattern recognition	Breath	[108]
Liver cirrhosis	Enose	VOCs pattern recognition	Breath	[108]
Asthma	Enose	VOCs pattern recognition	Breath	[108]
Diabetes		VOCs pattern recognition	Breath	[108]
Lung cancer	Dog	VOCs pattern recognition	Breath	[108]
Lung cancer	Enose	VOCs pattern recognition	Breath	[108,114]
Breast cancer	Dog	VOCs pattern recognition	Breath	[108]
Breast cancer	Enose	VOCs pattern recognition	Breath	[108,114]
Colorectal cancer	Enose	VOCs pattern recognition	Breath	[108,114]
Lung cancer	Proton Transfer Reaction Mass Spectrometry (PTR-MS)	Formaldehyde	Breath	[107]
Chronic renal failure	E-nose	VOCs pattern recognition	Breath	[107]
Alcoholic hepatitis (AH)	Selected Ion Flow Tube Mass Spectrometry (SIFT-MS)	2-propanol, acetaldehyde, acetone, ethanol, pentane, trimethylamine	Breath	[107]
Chronic Obstructive Pulmonary Disease (COPD)	E-nose	VOCs pattern recognition	Breath	[107]
Airways inflammation	E-nose	VOCs pattern recognition	Breath	[107]
Gastric Cancer	Sensors, Gas Chromatography Mass Spectroscopy (GC-MS)	6 discriminant VOCs	Breath	[107]

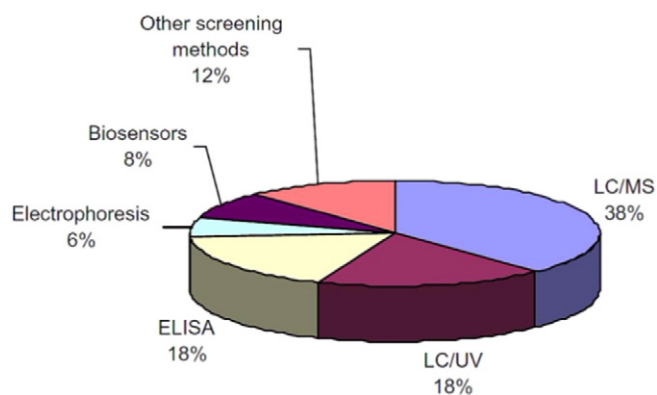


Fig. 1. Methods of screening antibiotics in food [19].

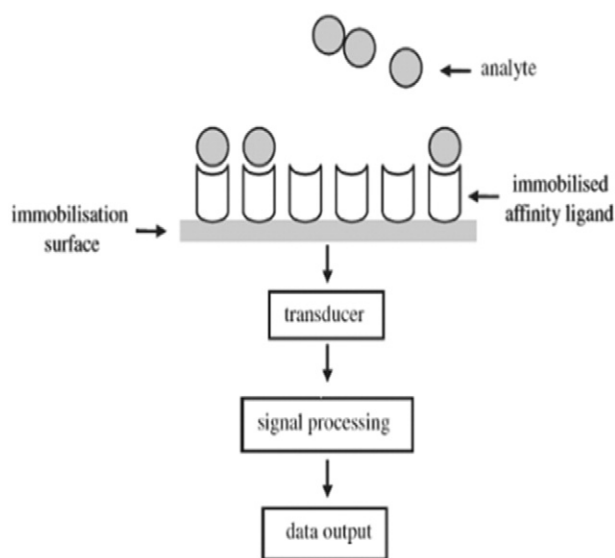


Fig. 2. Schematic representation of the configuration of a biosensor [19].

be measured by glucose and lactate. Intestinal ischemia causes variation of intracellular biochemical markers, which assists in the physiological monitoring of the bowel under stress. Online biosensors have been used successfully to monitor the effect of hypoxia on metabolic rates [103].

Monitoring the metabolism of free-moving animals can sometimes be an arduous task. However, an implantable device with a micro-fabricated sensing platform, a power coil and custom designed integrated circuits has been proposed. It allows the electrochemical detection of endogenous and exogenous metabolites. An epoxy-enhanced polyurethane membrane capable of retaining enzyme activity up to 35 days

was used for biocompatibility. Using a biocompatible membrane produced less inflammation at the site of implantation of device. Electrochemical sensors for glucose and lactate measurement in animal bodies are commercially available, and more such prototypes are being validated. Implantable devices for monitoring drugs would help personalize dosages of drugs for animals. Electrochemical sensors are being proved as ideal devices for rapid analyte detection and are helping us monitor the therapeutic range of various metabolites. The sensor sensitivity is improved by using nanostructured materials. The biocompatibility of sensors should, however, be addressed. Carbon nanotubes have proved to be advantageous in recent research, but they may be toxic. When implanting a sensor in animal body, the sensor stability must be kept in mind, so the host can tolerate the sensor and there is a limited foreign body reaction. Moreover, sufficient power should be supplied to these implanted devices, as heavy batteries need to be replaced and may add as an undesirable bulkiness to the device [104].

2.14. Breath analyzer

The composition of volatile organic compounds in the breath can provide deep insight about blood glucose level. The Volatile Organic Compounds (VOC)s are separated and quantified for this purpose. Usually, the glucose level in blood is associated with VOCs like ketone bodies, ethanol, methanol and exogenous compounds [105]. Moreover, volatile composition of the exhaled breath can be used for breath analysis—a noninvasive approach for urgent diagnosis. These diseases include many cardiovascular (CVDs) and chronic respiratory diseases. The volatile composition of breath reflects the composition of blood-stream and airways, which gives a comprehensive status of the organism's metabolism. Procedures like solid-phase and needle trap micro extraction can be combined with modern analytical technologies, such as mass spectrometry, which allows the analysis of exhaled breath. This facilitates the statistical analysis of heterogeneous datasets obtained from research and allows early disease diagnosis [106]. Scent detection from animals by electronic devices can be used for disease detection. Dogs and rats are frequently used for the detection of lung cancer and tuberculosis. Their noses have special sensory cells that detect the presence of certain volatile compounds. Man-made electronic devices, such as Enoses, has attempted to replicate this intricate biological system with moderate success [107].

Bovine tuberculosis is a cattle disease with an international public health importance. The ability to identify volatile organic compounds produced by pathogens has led to an emerging interest in human and veterinary medicine alike in diagnosing this disease. *M. bovis* infection can be easily identified through changes in the volatile organic compound profiles present in breath [108]. Many established breath biomarkers, like ammonia, methane, carbon dioxide, acetone and nitric oxide, can be analyzed by high-sensitivity laser spectroscopic techniques, like tunable diode laser absorption spectroscopy (TDLAS), integrated cavity output spectroscopy (ICOS), cavity leak out spectroscopy (CALOS), cavity enhanced absorption spectroscopy (CEAS), and quartz enhanced photoacoustic spectroscopy (QEPAS). Fingerprints of the

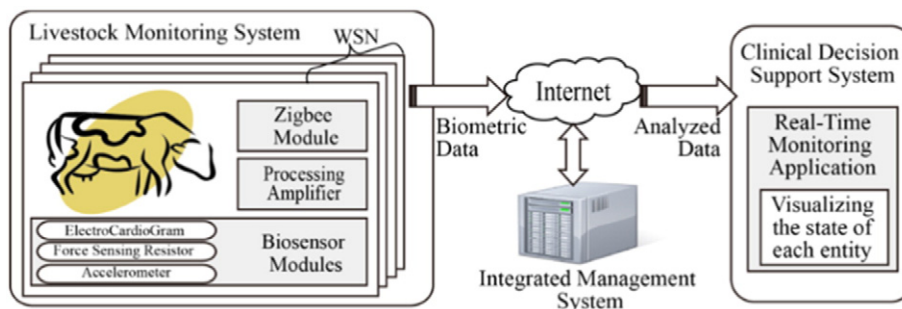


Fig. 3. Schematic of a Livestock monitoring system depicting the deployment of biosensors technologies [73].

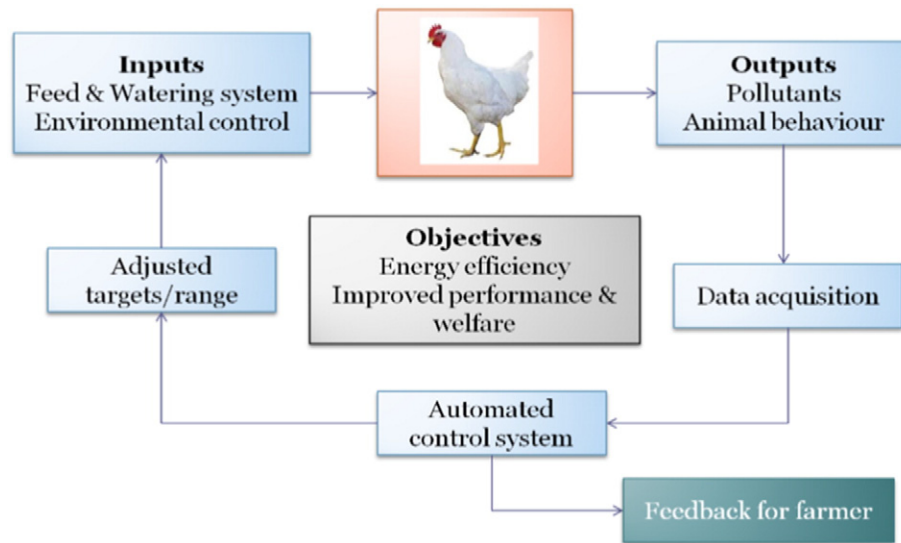


Fig. 4. Schematic of a health indicating parameters and biomarkers measured by biosensors in a Poultry farm [53].

biomarkers extend from UV to IR spectral regions. These sensors, using the laser spectroscopic techniques, are now commercially available and employed for breath analysis [109,110]. Gas-chromatography/mass-spectrometry analysis can reveal the presence of VOCs associated with *M. bovis* infection. A nanotechnology-based array of sensors has been tailored for detection of *M. bovis*-infected cattle via breath, which allows real-time cattle monitoring [111].

Sources of foot-and-mouth disease can be identified using a non-invasive general screening approach. This method employs hand-held air samplers using electrostatic particle capture, which captures airborne infectious agents and are later subjected to real-time PCR. This technique allows foot and mouth disease virus monitoring in epidemiological contingencies [112,113]. Arrays of monolayer-capped gold nanoparticle (GNP) sensors can be used effectively, in combination with pattern recognition methods, for fast and cost-effective diagnostic results from exhaled breath samples. The rationale and benefits of using breath analysis utilizing monolayer-capped GNP sensors in different fields of medicine like infectiology, respiratory medicine and oncology have been thoroughly determined [114,115]. A recent patent, which

may be helpful to determine respiratory health in animals, can detect carbon dioxide from fluid [75].

2.15. Digital animal health

Advances in digital technology are revolutionizing the animal health market. Apps have been developed to strengthen the relationship between veterinarians and pet owners. Engineers at all leading companies are developing digital technology to fulfill the needs of pet owners, dairy farmers and livestock managers. PetDialog is an app that allows users to monitor exercise and nutritional intake, socialization and other activities with the help of a built-in calendar that sends out alerts for routine care such as vaccines. Veterinary hospitals were found lacking in online appointment scheduling and online money transfers systems, and had cumbersome animal insurance procedures, but new development of digital systems is making it easier to manage animal health. Apps on smartphones can track and send animal behavior to veterinarians for faster, more accurate medical aid.

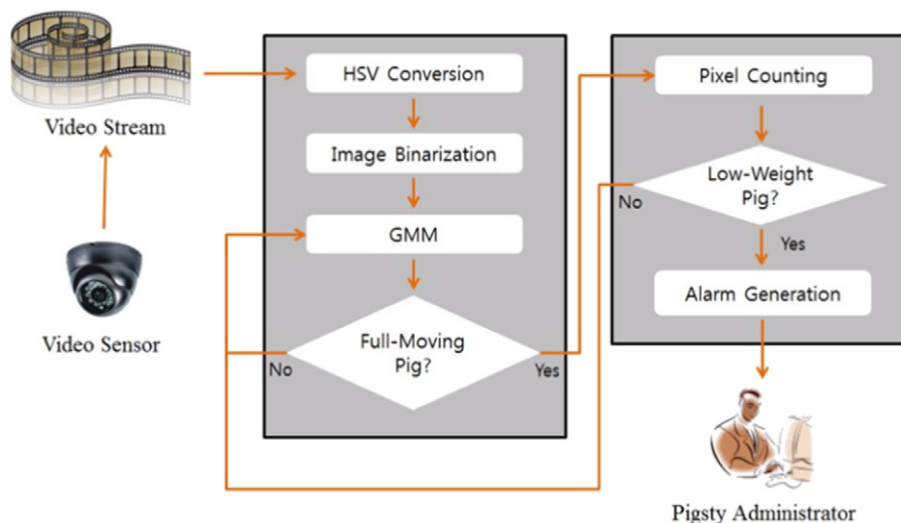


Fig. 5. Schematic showing a framework deploying sensor technologies for detecting low-weight pigs [9].

Apps are also being designed to automate long-standing practices in food production. An app called BCS Cowdication has been released in ten languages due to its ability to track cow's body conditions. Many illnesses that spread to humans originate in animals; therefore, digital health technologies can help control disease outbreaks [116].

2.16. Economic consequences

Several ingenious sensing devices and concepts have been demonstrated and also proposed in recent past years, but building a miniaturized device that can transmit data in a real-time fashion and also can simultaneously detect multiple target molecules remains a bottleneck.

Farmers must consider the economic dimensions of investing in sensors. The accounting data of farms can provide significant information regarding the productivity of these farms and the impact of using sensor systems. A study conducted on 217 Dutch dairy farms has proved to be useful in this regard, giving us insight into the economic consequences of such investments. The Malmquist Total Factor Productivity index is used to measure the productivity change for farms with and without sensor systems. This index reveals the gradual changes in a farm's productivity, and tells us how technical changes contribute to success. Recent studies have suggested that sensor systems may significantly influence the productivity on dairy farms, and this issue needs to be explored for economically viable deployment of sensors. Farmers should be briefed more about techniques to improve the performance of sensor systems, and the claims of the producers of such devices must materialize on farms [117].

3. Conclusions

Animal health is a serious global issue that demands apt scientific techniques. For this purpose, innovative approaches, like the use of biosensors for animal health management, has gained recognition. These sensors are at various steps of commercialization, but are making their way into the practical use and application in the domain of animal health. Some technologies for gaining an accurate health status and disease diagnosis are applicable only for humans. With modifications and testing in animal models, these innovative technologies are now being considered for their future use in livestock development and welfare. Precision livestock farming techniques, which include a wide span of technologies, are being applied, along with advanced technologies like microfluidics, sound analyzers, image detection techniques, sweat and saliva sensing, serodiagnosis and others. However, there is a need to integrate all the available sensors and create an efficient online monitoring system, so that animal health can be monitored in real time, without delay. Looking at an optimistic future of different wearable technologies for animals, including nano biosensors and advanced molecular biology diagnostic techniques for the detection of various infectious diseases of cattle, a large-scale adoption of the modern techniques discussed here is likely.

Author contributions

SN designed the concept of this paper, and critically reviewed and drafted the article.

Conflicts of interest

The authors declare no conflict of interest.

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