NONINVASIVE DIAGNOSTICS: PROGRESS AND CHALLENGES

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ABSTRACT

Non-invasive diagnostic procedures have become increasingly important in medical research and development because of their ease of use and low cost. It comprises integrated diagnostic gadget types, which are often regarded as the best way to streamline testing across healthcare systems. We explored recent advances and techniques in non-invasive diagnostics in this review, with an emphasis on tear, sweat, saliva, and urine-based biosensors. We have also discussed its application in diagnostics of lung cancer.

KEYWORDS: Biosensors, Diagnostic tools, Integrated devices, Nanotechnology, Polymer-based

INTRODUCTION

Non-invasive healthcare technologies have become more essential aspects of current research and development due to the cheap cost and convenience shared by both healthcare receivers and providers. Sweat (Olarte et al., 2013), urine (Morris et al., 1981), saliva (Aydin, 2007a), and tears are examples of non-invasive measures, however fluid-free technology can also be used. The last alternative is more appealing since it allows for quick and inexpensive tests with fluid-independent techniques, and the scientific community's worries about bio analyte concentrations in blood and other physiological fluids may be ignored. (Morris et al., 1981; Moyer et al., 2012) Many other non-invasive instruments have been developed, such as cardiovascular diagnostic systems, bioimpedance based scales, and even non-invasive blood analyzers to measure sentinel substances in blood, such as hemoglobin, oxygen, and glucose, in addition to traditional fluid-free non-invasive technologies that have been known and used for ages, such as electrocardiography. (Fye, 1994)

The main purpose of this paper is to present an overview of current progress and techniques in non-invasive diagnostics and to take a look at the challenges faced in the examination of new ways to improve.

NON-INVASIVE

Diseases, procedures, and devices which do not insert into the skin can be described as non-invasive. It is a procedure that does not require the insertion of an instrument into the body through the skin. Hearing aids, external splints, and casts are examples of non-invasive equipment.

NANOMEDICINE

By introducing nanostructured materials or devices such as nanodrug((Augstine et al., 2020) carriers, diagnostic probes, biosensors, microfluidic platforms, and medical imaging contrast agents, nanotechnology in medicine offers immense potential to change cancer detection and therapy(Augustine et al., 2020; Park, 2007). Nanomedicine techniques used to rely on tweaking the characteristics of well-established diagnostic and therapeutic methods or devices. With the help of supramolecular assembly of simpler components employing nanoscale engineering concepts, extremely efficient innovative therapeutic and diagnostic techniques have emerged in recent years (Augustine et al., 2021). Nanoparticles' particular features are also being used to detect and distinguish tumor cells from normal cells, as well as to provide targeted site-specific treatment (Chandrasekaran et al., 2021).

Nanomedicine's purpose is to use manufactured nanodevices and nanostructures for monitoring, controlling, and improvement of human biological systems at the molecular level which is a process known as *single-cell medicine* and has the goal of producing medical benefit. The incorporation of nanoscale technologies into medical practice will radically alter how we diagnose, treat, and prevent sickness. We'll begin diagnosing and treating diseases at the single-cell level, rather than, for example, at the organ level.

With the use of suitable nanoparticles, we can generate a three-dimensional image of tissue microanatomy, which will aid in mapping the spatiotemporal distribution of malignant cells. Such diagnostic gadgets can be relatively inexpensive and simple to use, allowing them to be used in cancer detection at an early stage. Early cancer identification, strategies aiming at enhancing the sensitivity and specificity of nanoplatforms for concurrent and mass analysis, and associated bio-marker detection are among the primary benefits currently being researched in nanotechnology-based cancer diagnostics. Thanks to advances in nanotechnology, nanoparticles can now be employed for non-invasive imaging of tumor tissues.

BIOSENSORS

Biosensors are frequently used as appealing alternatives to the big, expensive, and complex analytical instruments used in the healthcare industry. Many of these devices, which use optical, piezoelectrical, and chemical transducers to detect various analytes, have been produced over the years. Chemical sensors in particular have been increasingly popular in clinical medicine due to their great performance, portability, simplicity, and low cost. The majority of these sensors, however, necessitate blood samples for diagnosis. Such invasiveness is inconvenient for the patient and obstructs the data collection necessary for regular health monitoring. This is especially true for newborns and elderly individuals, who have a difficult time getting blood. In a variety of medical applications, continuous monitoring is critical. Optimal diabetes management, for example, necessitates frequent glucose concentration monitoring. Athletes, too, require ongoing evaluation of their fitness levels.

Pathogens in physiological fluids can be monitored regularly and in real time to aid in the early detection of a variety of disorders. Another instance in which continuous measurements are clearly important is drug efficacy tracking. In this sense, intrusive devices are limited since it is impractical to have constant access to the specified fluid samples, such as blood and urine. Sweat and exhaled breath gas sensors have received

a lot of interest in the last ten years. These nonintrusive sensors provide impressive performance for frequent or continuous health and fitness monitoring. It is feasible to change people's lifestyles by using the data provided by these sensors at regular intervals.

BIOSENSORS IN TEARS

Chemicals found in eye tears such as lactate, glucose, ascorbate, and neurotransmitters like dopamine and norepinephrine, have diagnostic potential. As a result, studying the composition of tear fluid can be used to identify many types of diseases and conditions, and a recent review provides a good overview of this. Glaucoma patients have lower-than-average tear neurotransmitter concentrations, and measurement of catecholamines in tears has been supported in glaucoma diagnosis. Furthermore, because the baseline concentration of the stress marker norepinephrine is high enough for detection, and because the concentration is anticipated to grow during psychological and physical stressors, a non-invasive tear-based stress sensor should be possible.

Tear sampling is an arduous task, and all the currently available methods have flaws. Tears can be absorbed using Schirmer strips on the lower eyelid, but the method tends to capture cellular as well as secretory proteins, and the physical presence of the strip can cause mechanical stimulation of the corneal and conjunctival epithelium, causing reflex tears to be released(Green-Church et al., 2008). As a result, the composition of the samples gathered thus far is unlikely to be the same as that of native basal secretion (Markoulli et al., 2011). While microcapillary tubes appear to be less invasive than Schirmer strips for drawing tears from the reservoir within the conjunctival sac, collecting basal tears with this method can be difficult and time consuming, and pooling samples may be essential to get sufficient volumes for analysis (Sack et al., 2007). Furthermore, the Schirmer strip reflex tear concerns are present in the microcapillary tube approach. To summarise, the little quantities collected combined with low concentrations of tear bioanalytes may account for the wide variations in concentration estimates reported, and it appears that authentic and correct bioanalyte concentrations in human tears are yet unknown.

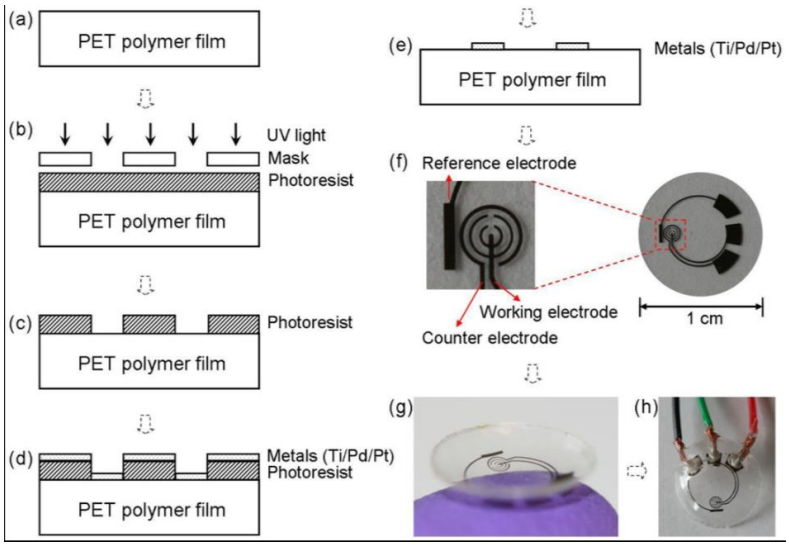


Figure 1 Sensor fabrication process(Jing Xu et. Al. 2021) along with its results

BIOSENSORS IN SWEAT

Biofluid sweat is used in other sensing technologies for the evaluation of numerous analytes (Chung et al., 2019; Nyein et al., 2016). Sweat contains several substances that can assist doctors figure out things like health and metabolic status, physiological state, and disease states (Munje et al., 2015; Nyein et al., 2016). Sweat glands are abundant on the outer skin surface and are densely and broadly dispersed throughout the body (Kim et al., 2019). Through microscale pores, these sweat glands generate and expel an acidic fluid straight to the outside skin surface (Chung et al., 2019; Corrie et al., 2015). Sweat also contains a variety of compounds such as lactate, glucose (Nyein et al., 2016), cortisol, testosterone Click or tap here to enter text., uric acid, and bigger molecules such as peptides, cytokines, and proteins. (Kim et al., 2019).

Furthermore, because of the link between blood analyte concentration and discharged sweat analyte concentration, analyte detection in sweat is possible (Karpova et al., 2020). A strong sweat-blood association has been shown for analytes such as hormones (cortisol, testosterone), potassium, and several substances, such as alcohol. Furthermore, a relationship between perspiration and blood glucose levels has been found, with the potential for use in diabetes monitoring (Chung et al., 2019). Although there is no established association between blood & sweat lactate or urea concentrations, actate/urea measurements in sweat can provide an indicator of the health status of the studied patient.

The sensor must be in close contact with the skin when collecting data from perspiration, ideally with a planar fit. Sweat biosensors are also commonly made with a flexible substrate for interaction with the skin's surface, allowing for a wearable sensor platform. Both planar fit and flexibility provide effective sweat sample collection and a smaller sample volume requirement (Bandodkar & Wang, 2014; Chung et al., 2019; Nyein et al., 2016). A rapid reaction time of the detected analyte, high stability, selectivity, and sensitivity under environmental conditions are additional requirements for a wearable sweat sensor (Bandodkar & Wang, 2014). Electrochemical readout methods, such as amperometry and electrochemical impedance spectroscopy, can be used to realize and achieve a quick sensor response time. The use of electrochemical methods in sweat biosensor sensor creation provides several benefits, including low cost, excellent performance, and device portability (Bandodkar et al., 2014). However, normalization of the sampled volume is a considerable disadvantage when undertaking quantitative analyses. Monitoring the sweat flow rate, for example, by measuring the change in sweat generation rate using skin impedance, is one technique to mitigate this. The sweat rate, however, does not predict actual biomarker sampling intervals without a comprehensive fluidic model between the sweat glands and sensors. (Falk et al., 2020)

Figure 2 Some examples of wearable(Jing Xu et al. 2021) sweat-based biosensors

BIOSENSORS IN SALIVA

Saliva is an oral fluid produced primarily by three pairs of major salivary glands: parotid (inside cheeks), sublingual (under the tongue), and submandibular (bottom of mouth), as well as a vast number of tiny salivary glands (Ilea et al., 2019; Malon et al., 2014; Segal & Wong, 2008). Saliva is also a clear, viscid, complex, colorless, and odorless fluid with a pH of 6.6–7.1 (Liu et al., 2019; Segal & Wong, 2008). Saliva is a watery material that contains bacteria, leukocytes, epithelial cells, crevicular fluid, hormones, ions, enzymes, proteins, nucleic acids, antimicrobial components, cytokines, and antibodies. These many components, which originate in the blood, might diffuse into the oral cavity via para-cellular or trans-cellular pathways, adding to the complexity of saliva (Bandodkar & Wang, 2014; Ilea et al., 2019).

The link between cortisol concentrations in saliva and blood has been demonstrated, allowing for the development of various biosensors based on their detection (Dorn et al., 2007; Mitchell et al., 2009). An NiO thin film-based label-free electrochemical immunosensor was used to create a highly sensitive and non-invasive electrochemical immunosensor for salivary cortisol detection. Cortisol antibodies were fixed on the electrode surface with EDC and NHS and detected by differential pulse voltammetry (Falk et al., n.d.). Interdigitated microelectrodes and anti-cortisol antibodies were covalently bonded on self-assembled monolayers of dithiobis to create another electrochemical label-free immunosensor (DTSP). Using cyclic voltammetry, different cortisol concentrations were assessed.

Saliva analysis offers a lot of potential when it comes to general health monitoring (Ilea et al., 2019). Saliva contains a diverse set of biomarkers that can be used for clinical analysis and illness diagnosis. Changes in saliva composition also reveal the current health status of the examined person because many biomarkers discovered in saliva are transmitted directly from the bloodstream. Different types of biomarkers and metabolites such as lactate, ethanol, cholesterol (Guilbault et al., 1995; Pappa et al., 2016), or glucose have been linked to blood and saliva concentrations. As a result, saliva analysis allows for the monitoring and surveillance of the human body's emotional, hormonal, nutritional, and metabolic states (Kim et al., 2019). Furthermore, using saliva as a type of diagnostic fluid has a number of advantages, including a painless and non-invasive technique for diagnostics and monitoring, as well as a simple and quick collection method. Furthermore, sample collection is simple (Ilea et al., 2019; Malon et al., 2014), does not invade the patient's privacy (Malon et al., 2014), and does not necessitate special laboratory equipment or skilled medical professionals. Furthermore, saliva can be utilised as a replacement fluid because it is generally safer and has a reduced risk of contamination than blood tests (Ilea et al., 2019; Kim et al., 2019; Malon et al., 2014). As a result, saliva can be used to assess a variety of biomarkers in a simple, reliable, and cost-effective manner (Kim et al., 2019) and can be used to perform multiplex metabolite detection with excellent sensitivity and selectivity (Pappa et al., 2016).

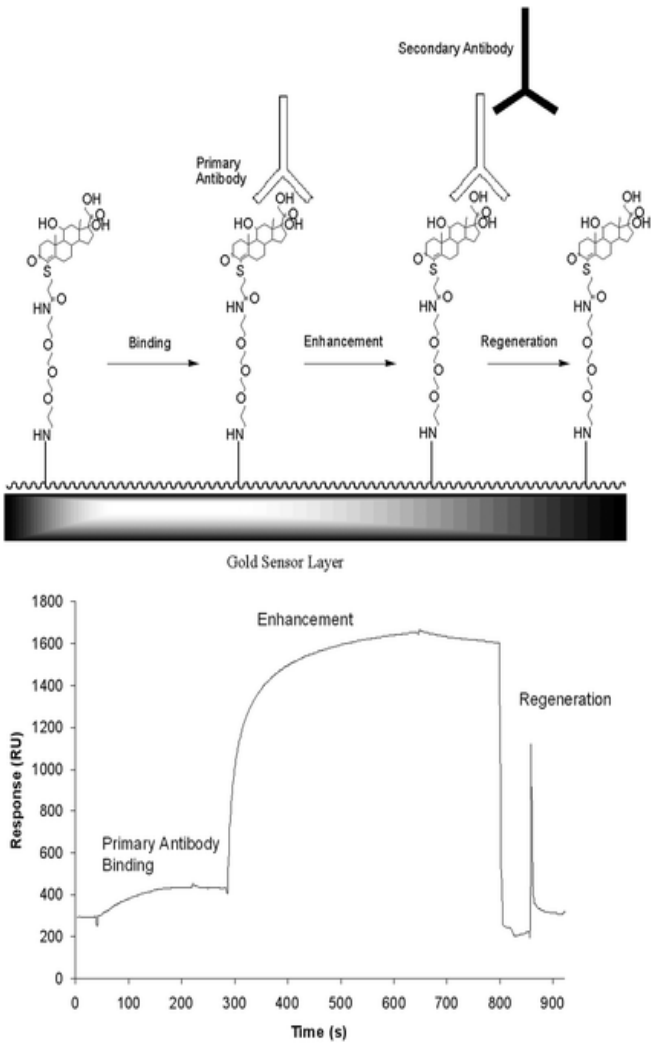


Figure 3 This is a schematic of the SPR biosensor cortisol assay(Jing Xu et. Al. 2020) along with a sensorgram plot of response ((RU)) vs. time (s)

BIOSENSORS IN URINE

One of the most well-known biosensors for detecting bio analytes in urine is oxidase-based biosensors. These sensors detect the H2O2 produced, allowing the concentration of the analyte to be estimated (Rocchitta et al., 2016). GOx (Wilson & Turner, 1992), as in other circumstances, is the most commonly utilized enzyme for detecting glucose in urine. The use of a conductometric bio transducer, which generates a binary response when the analyte is present in urine, is one of the approaches used to detect glucose in urine using amperometry biosensors. The working idea is based on a GOx-modified Prussian blue-cellulose acetate layer. H2O2 is generated when the substrate is present, reacting with the layer (Figure 4). The reaction causes the conductivity of the Prussian blue-cellulose acetate layer to vary, allowing a wireless biosensor to estimate the amount of glucose. Amperometry with redox mediators and a bi-enzyme system is another method for determining glucose in urine that has been reported. By compressing electrically conductive carbon with the strip of a biosensor containing two redox mediators, namely an enzyme system for glucose oxidation and a silver/silver chloride reference electrode, the measurement can be made. The

analytical readout can be done by putting a drop of urine on a sensor and comparing the result to a standard calibration curve, or by translating the current flow to urine glucose levels in some units.

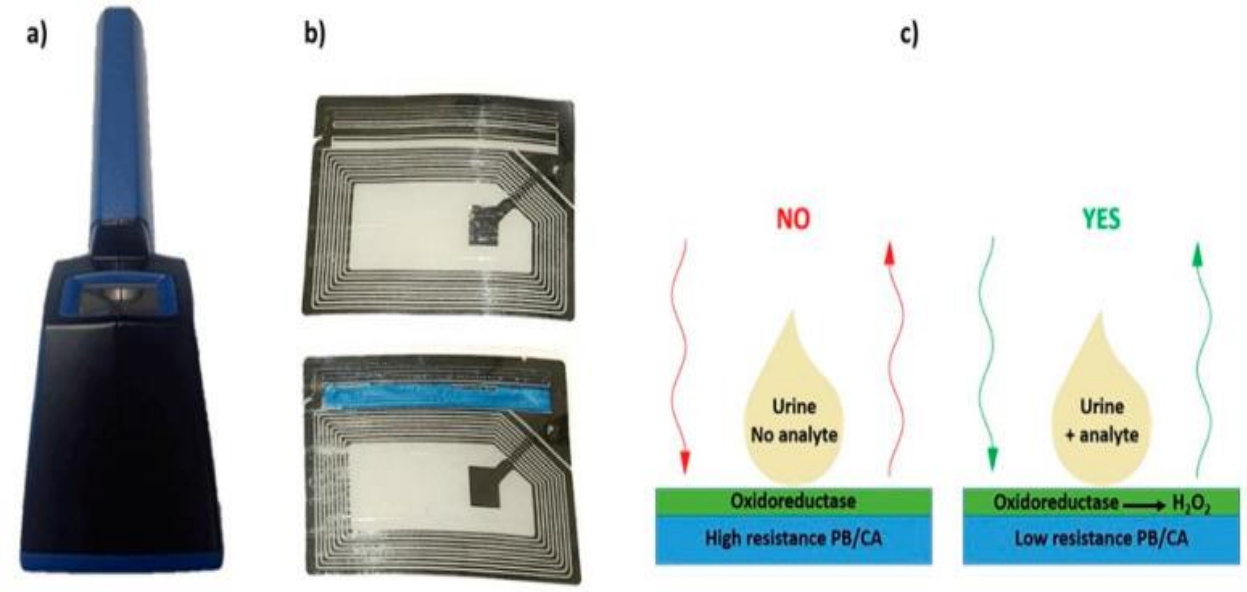


Figure 4 Moisture detection sensor circuit and other examples of urine-based biosensors (Jin Xu et al. 2021)

In the absence of microbiological identification at point-of-care facilities, physicians commonly commence broad-spectrum antibiotic treatment, contributing to the rise of resistant bacteria. Biosensors are the foundation of effective diagnostic tools for infectious diseases. The UTI Sensor Array (Mach et al., 2011; Pan et al., 2010) is an interesting example of a biosensor for uropathies identification. The sensor-platform is an electrochemical sensor array with bacteria-specific DNA probes as recognition elements. A self-assembled monolayer is applied to each of the 16 sensors, allowing for surface modification adaptability while also reducing background noise (Chaki & Vijayamohanan, 2002). A library of DNA probes targeting the most prevalent uropathies is immobilized on the sensor's surface (Liao et al., 2006). Hybridization events are converted into quantifiable electrochemical signals, which is how the detection process works.

Recent research has focused on developing wireless bacteria-sensitive biosensors based on near-field communication and radio frequency identification tags (Larpant et al., 2019). The methods were made possible by including a biosensor electrode inside the tag antenna. The wireless biosensors' transduction mechanism is based on biological redox reactions. The reactions alter the impedance of the tag antennas, which may subsequently be monitored wirelessly using a vector network analyzer or a cell phone. The future development of these wireless biosensor’s tags will focus on UTI monitoring as well as bacterial growth monitoring in hygiene and medical items.

INTEGRATED DEVICES WITH NON-INVASIVE SAMPLING

Here, we will discuss using eight different types of samples inside the context exploring the use of integrated testing devices and opportunity given by non-invasive sampling to connect directly to biologically relevant indicators with least amount of difficulty and supervision possible.

SWEAT

Sweat diagnostics have been a staple of physicians' toolkits, and they have recently gained much traction, especially in health monitoring. The prime driver of this interest is its relatively easy possible by means via extensively dispersed sweat glands throughout the organ. This makes it an ideal medium for non-invasive sensing. Nonetheless, its one-of-a-kind secretion process stymies the practical implementation of sweat-based sensors. The cystic fibrosis (CF) screening is the sole notable clinical success. Sweat from the eccrine glands' transports physiologically relevant indicators from the bloodstream to the skin's surface for repeatedly monitoring by electroosmosis. Within the sweat duct, it causes a pulsatile advective flow. This discrete transport system causes a time lag and temporal inconsistency. Sweat-based analysis has a discontinuity compared to blood/interstitial fluid, making it difficult to achieve quick results.

The tight protein junctions along the course of the analyte transport is another concern. These proteins filter big biomarkers and effectively limit their amount along the cell membrane pathway, acting as a transport barrier. The changing nature of sweat production is not clearly realized for most analytes, so this needs to be resolved before sweat-based sensors can excel. Problem is replicated in the available research and commercial systems, which tend to focus on fitness markers like electrolyte concentration and sweat rate of loss instead of overall fitness.

TEAR FLUID

Proteins, hormones, electrolytes, nucleic acids, and glucose are among the ions and tiny molecules that can penetrate tears via the blood-tear barrier. The link between the blood and tear components is based on this diffusion mechanism. It also works as a transit barrier, allowing for easier sample analysis by differentiating blood cells and minimizing the matrix impact. However, as compared to their blood levels, this transport resistance lowers analyte concentrations. The continuous release mechanism of tears, which changes both the tear contents and their levels, is another prominent problem. Because it is always direct contact across the barrier membrane, basal tear fluid, which creates the permanent protective layer casing the eye, is the most suitable for analyte regulation. This discrepancy adds another element to the equation i.e., response tear flow pace and composition, further complicating the already complex blood-tear relationship. Microcapillary tubes can be used to directly collect basal tears as an alternative. In this situation, however,

specialized workers are necessary to prevent ocular damage, and the sample amount that may be collected is small (5 microliters).

Even after overcoming this technical hurdle, a greater difficulty awaits in-vivo testing of the created system on human participants, preferably incorporating testing process tests for the tear-blood relation. At last, all these research should yield an expense product.

SALIVA

Saliva provides a simple yet sophisticated non-invasive method for measuring the blood concentrations of a variety of analytes that can be used to track a patient's immunological status. Its complexity stems from the fact that it is made up of protein secretions from different glands compacted epithelial cells and microfauna, blood-derived compounds. The existence of a signature, and its degree, is influenced by both the sample collection strategy (whether it is stimulated or not) and the sampling method. Moreover, biomarkers' basic characteristics alter the compound's durability in its native environment, necessitating sample well before.

On either hand, technology has made it easier overcome these challenges by presenting an only one, specimen technique that has been effectively deployed as an on-sensing platforms or handheld devices.

Despite all the positive advancements, there are still many issues to be tackled. Identifying the transport mechanisms correctly is the most difficult task.

i) their unique physicochemical characteristics

ii) active / passive mechanism for movement

iii) contamination due to smoking or alcohol

iv) There is also bias because of oral medications and proteolytic enzymes

v) Breakdown during the blood-to-saliva transfer

URINE

Urine has been utilized as a diagnostic and therapeutic instrument for diabetes, infectious diseases, renal diseases, hydration, pregnancy, or even genitourinary cancer from the start of time. The creation of urine allows for the passage of metabolites, and other relevant sample matrix from adjacent blood capillaries, resulting in a realistic method for measuring biomarker concentrations that reflect the patient's condition. This diffusion method, like some other non-invasive media, results in substantially lower analyte concentrations; hence, only someone with an elevated plasma level is identifiable in urine tests if extremely reliable detection technologies are used.

Commercially accessible integrated sensors are being used to keep track of illnesses that are infectious. They are also used to analyze urine chemistry by detecting a variety of critical factors. However, due to

differences in analysis of color by end-users, these goods may cause test findings to be misinterpreted. To address this issue, firms have developed optical readers like as the Roche Urisys and the NephroCheck Astute140 Meter. The limited sensitivity of immunochromatographic techniques is another issue with them. In this regard, commercial fluorescent-based solutions have benefitted appeal as an acuitysolution, particularly for the detection of renal damage.

STOOL

In clinical settings, stools are checked on a regular basis, specifically in the context of gastrointestinal system diseases.. Its rich contents, which include many biomarkers, cells, germs and some parasites, contribute to its enormous potential for integrated sensors. Stool cultures are used to diagnose pathogenic disorders like dysentery and cancers of various sorts. Furthermore, the gut microbiota is linked to neurological illnesses such as Alzheimer's disease. Traditional stool analysis, in comparison to other non-invasive approaches, necessitates extensive sample processing, sophisticated equipment, and well-trained workers. Its time-consuming and labor-intensive nature further limits its use in the healthcare system. Another concern with faeces management arises as a result of the restricted access to the laboratory. Because the material is biohazardous, specific precautions must be taken during sampling and transportation. Furthermore, depending on the analyte, the period between sample collection and analysis can affect test results.

For Helicobacter pylori identification, crude stool samples are processed directly using a microfluidic device. The sample is first mixed using magnetic beads, that capture the DNA of the bacterium. After that, the beads are transported to an elution chamber, in which the DNA is cleaned before further analysis. Stool samples are homogenized in another investigation utilizing from the initial oscillatory flow, a subsequent consistent stream produced in a technology which processes on small number of fluids.

BREATH

Patients with flu-like symptoms were enrolled in pilot research at primary care centres in Belgium, and their diagnosis was established using nasopharyngeal (NP) swabs. After a very light process consisting of five typical exhales, experts collected expelled particulate from people with the disease using a breath electrostatic sampler (BESS) and filters. Quantitative Reverse Transcription PCR panel test was used to evaluate the samples. Design of sensor diagnostics can be done in two processes: choosing which biomarker must be investigated in a specific disease and how to detect that analyte. As a first stage, biomarkers in breath were analyzed, revealing some key VOCs which are closely linked to cell metabolism. In this context, breath analysis has been used to diagnose the disease. For diabetes, the acetone level in a person's breath is tested, and it's usually found to be at least two times greater than in a healthy individual. The ratio of specific VOCs, rather than their quantities, is used to detect lung cancer.

Low biomarker concentrations are the source of all the issues in breath analysis. The concentration of the trace chemicals in the moment is highly influenced by the subject's age, gender, food, and as well as pumping rate. Furthermore, the presence of exogenous VOCs can compromise a breath sample. The sample

collecting procedure, for example, can impact the composition, as the Infection may occur as a result of contact with saliva.

Because electrostatic tests have a lesser infiltrative speed and pressure loss than impaction devices, bacteria may be able to keep their purity. Electrostatic samples have also exhibited a 5–10 times higher concentration output than liquid bubblers. This device has the potential to be commercialized as an additional sampling technique for expelled breath particles in clinical settings. But right now, there is no technique that has been created to take samples from direct exhaled breath.

SEMINAL FLUID

Semen is a medium for cells that comprises sperm which contain protein, enzymes, vitamins and sugar. This is focused on measuring physical attributes and is the cornerstone of hormonal tests. In addition to sperm deficiencies such as state density, vitality, motility, DNA integrity, and shape, there are other factors to consider. In order to diagnose STDs, sperm testing is also required. CASA (computer assisted sperm analysis) and integrity testing are all examples of traditional clinical procedures.

Unprocessed, crude semen samples are analyzed using a device which pulled the fluid into microchannel to determine counting of sperm and his ability to move around. This method was put to the test by comparing its predictions with CASA, which revealed a better understanding. Analyzer based on smartphone was recently updated to include additional features for analyzing live sperm in sample of semen, and Separate pieces of DNA strand.

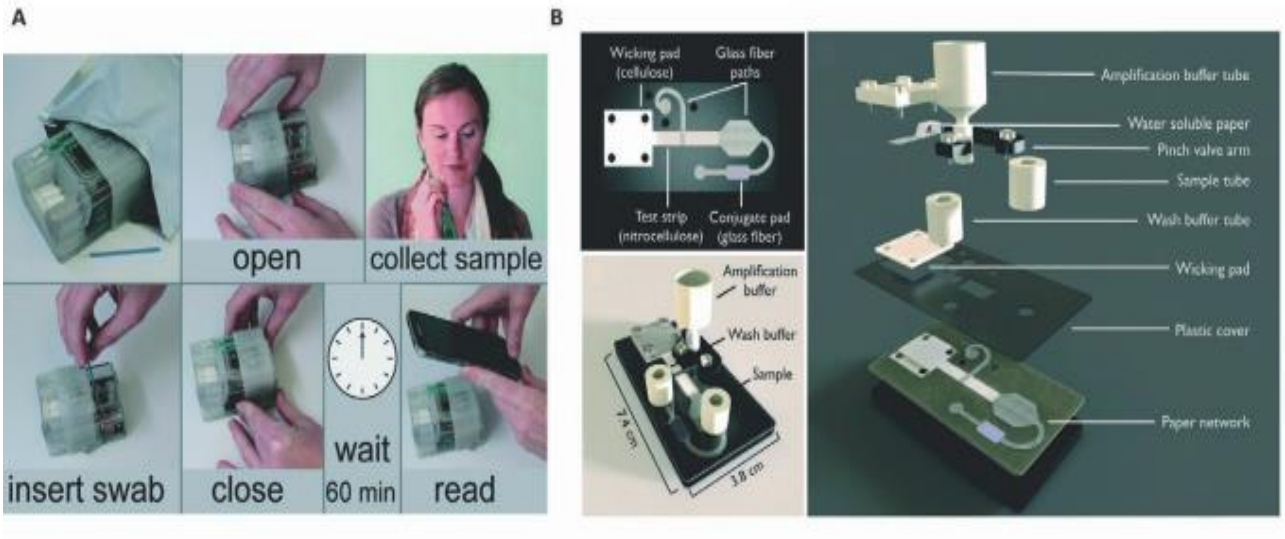


Fig.7 A) Diagnosing genes of MRSA bacteria from nasal
Fig.7 B) Diagnosing of influenza from nasal using 2D paper network

SWAB SAMPLING

Taking samples of swabs is a frequent and cost-effective diagnostic procedure for diseases. We can get samples of swab from nose, vagina, wounds. The most important stage in swab analysis is proper specimen collection, which includes sterilization, specimen processing, and handling of specimens. Swab must be flexible, form, and size appropriate for the application site, and the tip material must have the required qualities to enable efficient sample capture and target release. This method is useful in epidemic outbreaks such as COVID-19 to collect samples and which detect early of disease.

	Analyte	**Technology**
Stool	DNA	Electrochemical
Urine	Ascorbic acid, uric acid	Smartphone-assisted electrochemical
Swab	Hemagglutinin	LFA
Sweat	Glucose	Electrochemical
Breath	VOCs	Electronic nose
Tears	Glucose	Electrochemical
Semen	Concentration and motility	Smartphone-assisted optical
Oral fluid	Sodium	Electrochemical

Table 1

MINIMALLY INVASIVE TECHNIQUES

While non-invasive techniques do not involve tools that break or physically enter the body, minimally invasive techniques involve small incisions and a few cuts (one or more). This allows less pain, fewer complications (than invasive surgeries), and quick healing.

One of the first types of minimally invasive surgery was laparoscopy, which comprises surgery through one or more small incisions and the use of small tubes, tiny cameras, and surgical instruments. Another type of MIS is robotic surgery. It gives the surgeon an enlarged, three-dimensional image of the surgical site, allowing him or her to work with greater precision, flexibility, and control.

Minimally invasive surgery, abbreviated as MIS, persistently plays a crucial function in general surgery as a substitute to various standard laparoscopic procedures and open surgeries. Surgical procedures in MIS have significantly grown in popularity since the 1980s, thanks to technical improvement and creativity.

GROWTH OF MIS TECHNIQUES

To supplement MIS, the first growth pattern observed was in connection to surgical equipment and sutures. Surgical robots and image guidance are the second growth phase identified.

Despite various complicated engineering challenges, these technologies have shown continuing progress to keep up with clinical demand. Third-generation surgical robots are the outcome of continued robotic technology advancement. This may be seen in the rising use of robotics in numerous processes, with robotics sometimes even serving as a supplement to established methods like SILS. The most recent and third growth phase about NOTES I.e., natural orifice transluminal endoscopic surgery and SILS I.e., single incision laparoscopic surgery was observed in the late 2000s, with its onset in the mid-2000s and peaked shortly after. Though NOTES gained fame in late 2000s, SILS has grown in significance. (Raffaelli et al., 2019; Siddaiah-Subramanya et al., 2017)

Types of MIS:

1. Adrenalectomy, where one or both adrenal glands are removed
2. Brain surgery
3. Nephrectomy (kidney removal)
4. Colectomy, where parts of a diseased colon are removed
5. Heart surgery
6. Hiatal hernia repair, or anti-reflux surgery, relieving gastroesophageal reflux disease (GERD)
7. Kidney transplant
8. Spine surgery
9. Splenectomy to remove the spleen
10. Gallbladder surgery (cholecystectomy) relieving pain caused by gallstones

EVOLUTION OF SENSORS

Sensors have started to become one of the biggest and fastest-growing markets and are found everywhere compared to other technologies.

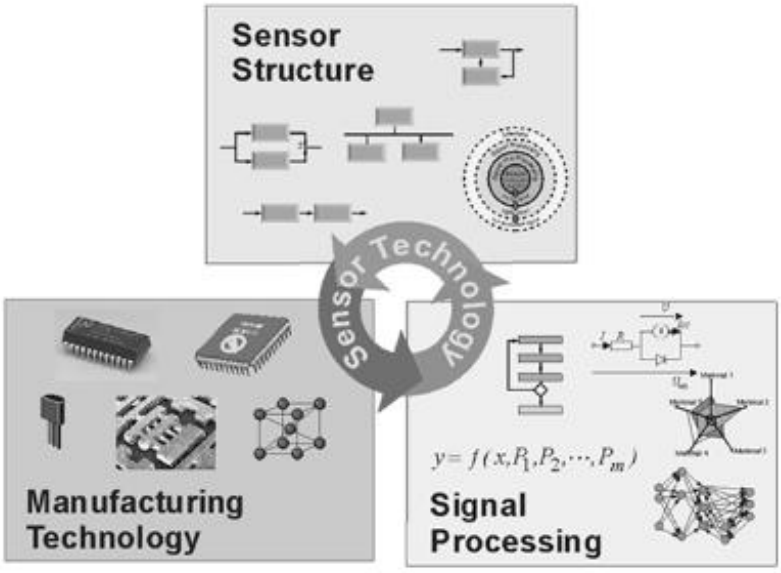


Figure 8 Sensor system depiction (Kanoun & Tränkler, 2004)

Sensors and sensor systems work by combining sensor structure, manufacturing technologies, and signal processing algorithms to perform their functions. Sensor systems are being used in whole new ways as the demand for automation, security, and comfort grows. As a result, advancements in sensor technology are dependent on continuous technical improvement in these sectors. New and innovative approaches to enhance sensor properties are used in digital signal processing. Calibration and consideration of a variety of impacts, like variation in the cross-sensitivity or manufacturing, becomes a straightforward procedure. Incorporating various other functions, like online self-calibration or self-test, is gaining special importance in improving quite much system reliability, and reducing maintenance costs & installation charges. (Kanoun & Tränkler, 2004; Trigona et al., 2020a)

There are various kinds of sensors used for different kinds of sensing. Some examples are temperature, humidity, infrared, ultrasound, light, smoke, alcohol, color sensors. We can also classify sensors into active and passive sensors where the former requires an external excitation signal while the latter does not. Another classification of sensors can be based on its conversion phenomenon, i.e., electro-chemical, photo-electric, electro-magnetic conversions. Based on the output signal sensors are classified as analogue and digital.

SENSOR TECHNOLOGY

The credit for recent advancements in technology goes to the development of micro technology.

These innovative technologies give the opportunity to manufacture systems with small dimensions, relatively low power consumption with high reliability in high volumes. Hence, microsystems now assemble sensors, actuators, mechanical, and electronic units. Like any other technology, there exist unique challenges for device modelling, its microfabrication and then packaging. Today almost all systems use micro machined systems which have become a very integral part of those devices like mobile phones, color printers, automobile and medical systems.

Being one of the remarkable Micro Technologies, Silicon micromachining reaps the properties of the silicon material, like least hysteresis errors. Prominent advancement in the field of microelectronics has paved the way to this important technical evolution. Dry and wet etching processes are used in bulk micromachining and its advantages are high etching selectivity and reliability. High sensitivity, displacement, mechanical strength, and reduced noise are hence the result of these properties. In the case of surface micromachining, three-dimensional mechanical structures are developed by a sequential deposition and selectively removing of sacrificial layers like SiO that separate the individual layers in the structure. (Kanoun & Tränkler, 2004)

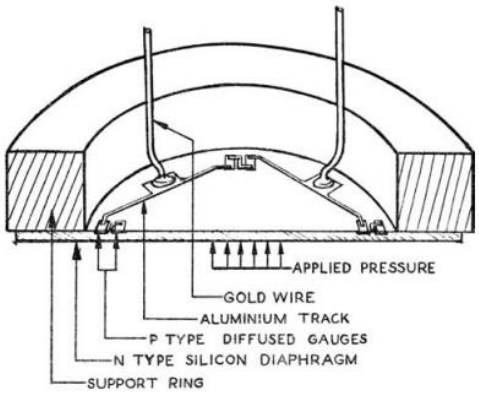
Focusing our attention over the past decade, the most prominent changes can be found in the adoption of technologies and processes ranging from centimeter-scale systems to millimeter-scale system devices. to silicon base, micromachining systems (MEMS) and nanoscale approaches (NEMS). Other polymers are materials capable of converting mechanical strains into electrical signals, in particular, electrically active ionic polymers (IEAP). The subject of different articles has been the combination of electromechanical conversion characteristics with the intriguing properties of lightness and elasticity.(Trigona et al., 2020b)

The most studied IEAPs are made with Nafion ®, which is a polymeric ionomer that is deemed harmful to the environment due to its chemical composition having fluorine. The highlight of this evolution is that it's biased towards green, biodegradable, and non-toxic environmental solutions. This appealing trait has been followed through novel materials possessing lead-free properties, the absence of hazardous metals, bacterial cellulose, that has been presented to be a feasible and environmentally friendly molecule for sensor fabrication.

MEMS-BASED SENSORS

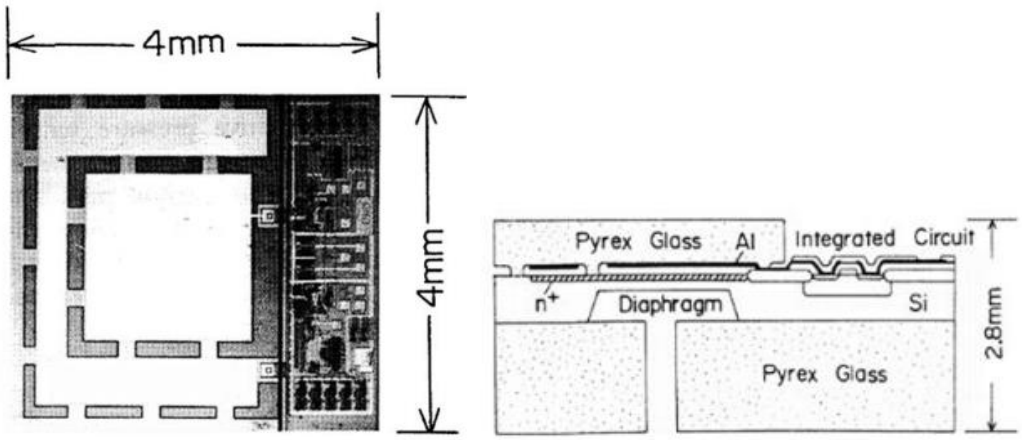
What is MEMS? The term is an imprecise designation because all MEMS devices are not electro-mechanical and only a small fraction are systems. However, it is now commonly used to describe a wide range of miniature devices, most of which are arbitrary 3D microstructures and fabricated from silicon using commonly discovered techniques. Manufacturing of integrated circuits is a complicated process where processes such as isotropic and anisotropic etching, different thin-film deposition methods, anodic bonding as well as masking and doping techniques are used. MEMS devices also include silicon microsensors, "lab on a chip" devices, and microparticle analysis systems devices. (Taymanov et al., 2014)

In the field of physical sensing, MEMS technology has delivered a variety of compact, durable, and inexpensive devices such as accelerometers, strain gauges, microphones, air mass flow sensors, pressure sensors, and, more recently, gyroscopes and yaw-rate sensors.



Note: The planar (non-microengineered) diaphragm mounted on the support ring and diffused rather than implanted "gauges" (piezoresistors)

Figure 9 Section of a silicon pressure sensor



(A) *(B)*

Figure 10 Latest MEMS pressure sensor with integrated electronics (A) Enlarged image of Sensor (B) Schematic cross-section

Today's pressure sensors are extremely miniaturized and reap the benefits of both capacitive and piezoresistive effects (where electrical resistance changes with change in applied stress). They are hence doped by ion implantation method instead of diffusion and also use non-planar diaphragms; and, after the 1980s, several sensors incorporated on-chip simultaneous signal processing and digital outputs. (Figure 10)

One of the most elegant MEMS sensors that has not yet been mass-produced are the yaw-rate sensors with complex geometries. Many sensors today have on-chip electronics, over-range stops, and self-test functions. (Bogue, Robert. 2007)

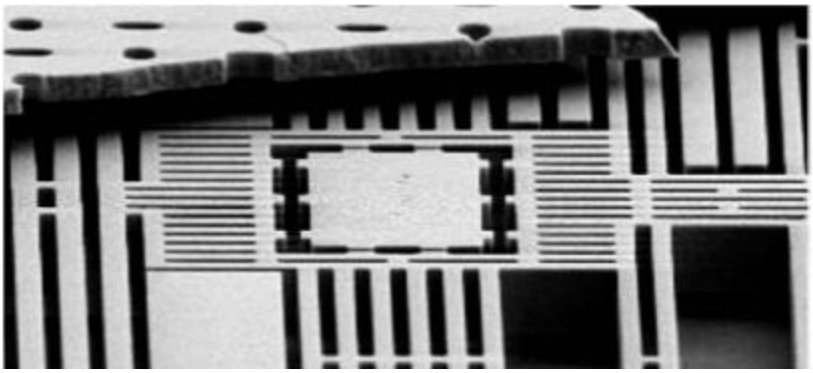


Figure 11 Automotive yaw-rate sensor showing complex MEMS microstructure

However, while in comparison to the complexity of Texas Instruments' digital micro mirror devices (DMDs) yaw-rate sensors are less complex. Although they cannot be classified as sensors, they are greatly recognized as the world's most intricate MEMS products. Despite 30 years of development, DMDs have not yet fulfilled their early affirmations and are now only suitable for hollow applications. The use of micro-engineered silicon substrates in metal oxide gas sensors, also called "micro hot plates," is the most common and modern MEMS gas sensing application. These are fairly straight forward devices, but a variety of more complex MEMS-based gas sensors and micro-spectrometers based on technologies such as NDIR (non-dispersive IR absorption), thermal conductivity, and photo acoustic are limited these days. However, it has been commercially successful. However, while such devices are compact, durable, and potentially inexpensive, smaller sizes often result in poor performance. Despite being used in one of the Viking missions to Mars, the commercial success of the "micro-GC" (or Miss) is limited to the study of various additional small analytical instruments for gases and liquids. It is paving the way for a-chip or micro-TAS.

POLYMER-BASED SENSORS

Conjugated polymers, after their discovery in 1960s, have been used in sensing technology for a wide variety of applications. This is due in part to the development of more sensitive sensors with lower detection limits and greater dynamic ranges. Advances in specifications and detection speed also contributed. Low cost, disposability, chemical, and electrical properties and tunability, and a well-defined interface between the conjugated polymer and the medium in which the analyte is present are all advantages of conjugated polymers for use in sensing applications. The ability to dope conjugated polymers provides benefits not just in terms of adding recognition features, but also in terms of allowing for further tuning and specificity.

Polymer-based sensing technologies are more promising than any other sensors for improving the performance of sensors and biosensors. Molecularly imprinted polymers (MIP), hydrogels, conducting polymers along with their composites, and other polymeric materials are employed in sensor devices.

Polymer-based materials are preferably used in such sensors as they increase target molecule recognition, acting as supports for immobilization of functionalities (e.g., dyes, fluorophores, metal), therefore allowing the detection of target analytes by altering their physical or chemical properties. Polymer-based sensors also have the advantage of being able to tune their chemical properties, such as reactivity, biocompatibility, flexibility, and resistance to degradation.

MORE ON SWEAT BIOSENSORS

These non-invasive devices enable real-time sweat analysis as well as provide useful information on a variety of health issues. Commercially available wearable sensors can only monitor a person's physical activity and other vital signs, they cannot provide relevant information at the molecular level. These are wearable biosensors that help in the monitoring of several analytes during physical activities, such as electrolytes, heavy metals, and metabolites.

Sweat biosensors, which provide important information on sports performance based on the quantity of water eaten, are used to monitor dehydration. Dehydration tests revealed a substantial increase in sweat [Na+] when individuals were given no water for 80 minutes.

Dehydration trials, in which participants were given no water for 80 minutes after dropping a pound,

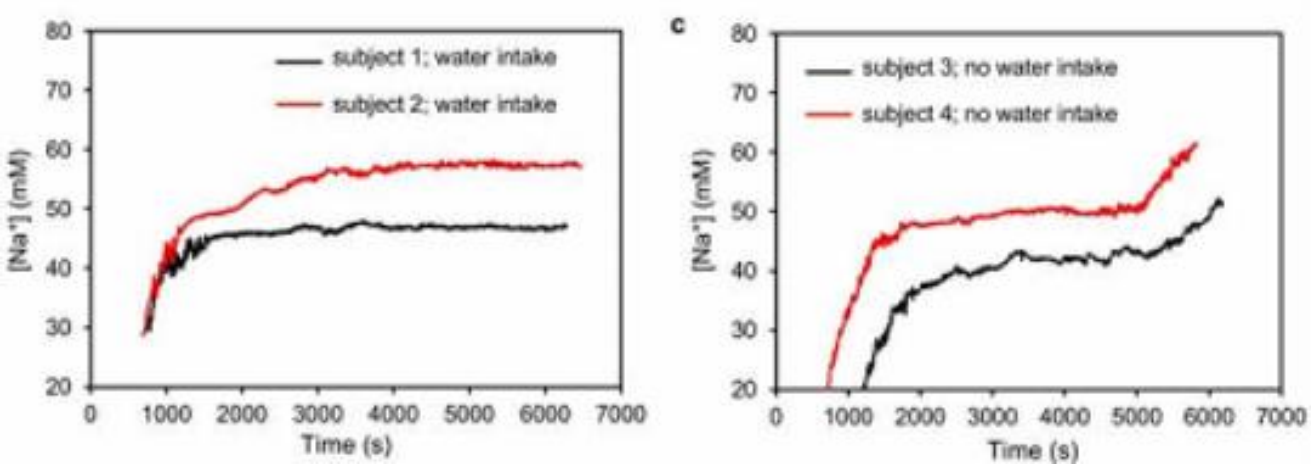


Figure 12 Water intake vs time graph

Sweat biosensors might be used to measure the amount and composition of heavy metals in the body, which is linked to human health. Sweat sensors that are printed & tattoo-based are increasingly employed for Zn monitoring. Stripping voltammetry has lately been used to test a variety of heavy metals, including Cu, Hg, Pb, Zn and Cd.

Wearable sweat sensors track major electrolytes like Na+ and K+, as well as metabolites like glucose and lactate, in real-time. The graph demonstrates that when perspiration rises, both glucose & lactate levels in sweat fall, signaling that the activity continues because of the dilution effect caused by the highly increased sweat rate. Initially, Na+ rises, while K+ falls.

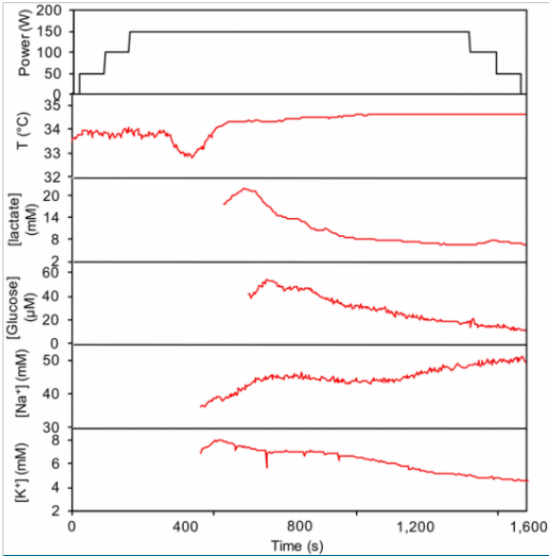


Figure 13 Ionic concentration vs time

In the end, pH is what decides how an illness is diagnosed. Calcium has an important role in human metabolism. Optical and electrochemical measurements of sweat pH sensors have been made. A completely integrated wearable sensor system for the simultaneous pH & Ca2+ evaluation in human sweat has also been demonstrated, which is especially important considering that free Ca2+ levels in biofluids are pH dependent. The graphic depicts a person's sweat profiles during a continuous load cycling activity: sweat pH rises dramatically for 5 minutes before stabilizing for the duration of the cycling workout, yet the Ca2+ sensor shows the opposite trend. A commercial and readily available pH meter which could be inductively coupled plasma-mass spectrometry can be used to validate the responses of these sensors.

High blood alcohol levels can lead to serious car accidents and higher cancer risks, whereas sweat ethanol levels are said to be similar to blood alcoholic levels (BAC). Wearable sweat ethanol sensors have been created, which combine iontophoresis based sweat or body fluid extraction technology with a flexible wireless circuit board. These sensors show distinct changes in current responsiveness before and thereafter consuming much alcohol, indicating a rise in blood ethanol levels.

Micro-invasive surgical tools:

Minimally invasive surgery is one of the most exciting and quite rapidly developing field or discipline where force sensing is important (MIS). We can perform the integration of micromachined piezoelectric sensors along with MIS equipment to improve diagnostic & treatment along with monitoring. A micro-machined freestanding lead zirconate titanate (PZT) force sensor was created using a five-mask method that included deep reactive ion, ion beam, and wet-chemical etching procedures. A 1-m thick PZT film is sandwiched between the top (Cr/Au) & bottom (Ti/Pt) metal electrodes arranged on a thin Si membrane in the PZT sensor, which is a parallel plate capacitor. The etch process provides effective method of etch control, helps to reduce undercut, keeps the photoresist mask in place, & successfully removes residues from etched surfaces. A fast etch rate (200 mm/min), strong photoresist selectivity, and little under-cutting (1.5:1, lateral: thickness). The manufactured force sensor's ferroelectric properties should be in excellent working order. The current manufacturing process and electrical analysis may be considered a milestone in

the fabrication of freestanding PZT force sensors in any desired shape and size, as well as a great example of ferroelectric microdevices.

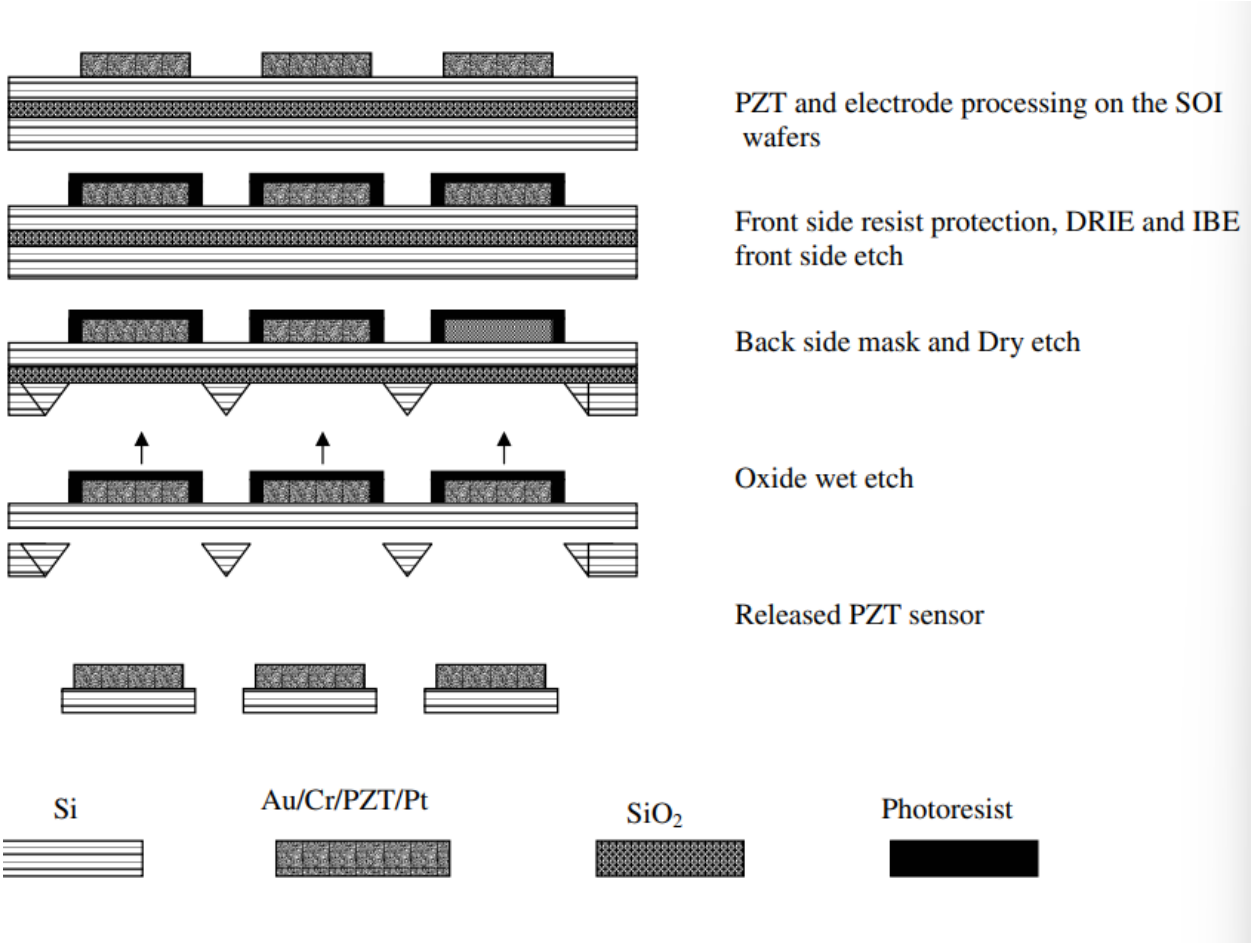


Figure 14 PZT (Force sensor release process) Fabrication sequence

Design and Fabrication:

PZT structures for thin-film and thick membranes are supported in the form of some cantilever structures with one or more than one ends on a micro-machined Si membranes or Si substrate. That is, all of these structures are permanently attached or clanged to the substrate, & the effect of the clamping of substrate on distorting the modes of displacements that cannot be evaded or avoided. In the tactile sensor applications, PZT force sensors should be fully free of the substrate, and the freestanding PZT force sensor leads the way for design flexibility and maximum force sensitivity in the mN to μN range. Device features of PZT-based microsensors and micro actuators used in biomedical MIS tools are very sensitive to the microfabrication process. The process of etching the PZT thin sheet is crucial to the device's success. Among the several dry etching processes, ion beam etching (IBE) is the most extensively utilized for designing PZT thin films without any undercutting. Due to the disparities in sputtering efficiency of Zr, Ti & Pb, as well as the low selectivity of PZT when compared to photoresist (mask) & Pt, obtaining appropriate PZT patterns is difficult (electrode). In reactive ion etching (RIE) procedures, PZT's etch selectivity over photoresist and Pt is equally poor, and it's difficult to make consistent PZT patterns even with high-density & plasma-assisted techniques like inductively coupled plasma (ICP) & electron cyclotron resonance (ECR). The previously mentioned processes are best for films less than 1 micron thick because of the slow etch rate (100-320/min). These techniques, however, become more complicated for thicker films.

Wet etching is a more useul and effective approach for trending MEMS than dry etching because of its high rate of etching, good selectivity and cheap cost.

Experiments:

Researchers attempted to put force sensors on the gripper jaw, where they would not be impacted by friction or other interfering forces, to directly quantify the force that is acting on the tip of the surgical tool. Although a 2 mm x 2 mm piezoresistive sensor for force measurement of grabbing the head was developed and achieved detection resolution of 0.1 N, where crosstalk remained in each and every direction, and sensor properties were impacted by temperature. A new capacitive transduction-based force sensor was described, which effectively delivered two-DOF force information, although sterilizing, assembly, and miniaturization were not considered. Complex packaging and electromagnetic interference are obstacles for electrical-based force sensors. As a result, optical fiber sensors have been used in surgical equipment by several researchers. Optical FBGs were used to measure the grabbing force on a surgical grasper, and the resolution of the clutching force reached 11 mN. (Spehar-Délèze et al., 2021) However, to compensate for the thermal drift which is of axial force sensing, the measuring system required a temperature compensation grating.

Nanorobotics:

Nanorobotics, which has long been a science fiction fantasy, has become a reality because of significant advances in domains such as physics, chemistry, materials, information, & nanotechnology in recent decades. Not only are many prototypes of nanorobots with nanoscale sizes created for diverse biomedical purposes, but also robotic nano manipulators capable of handling nano-objects gain significant breakthroughs for biomedical applications. The remarkable & exceptional achievements in nanorobotics have significantly expanded and contributed in the field of medical robotics and yielded novel insights into the underlying mechanisms guiding life activities, miraculously demonstrating an emerging and promising way for advancing diagnosis and treatment levels in the upcoming era of personalized precision medicine.

Type of nanorobotics		Overall size	Description
Nanorobot	Molecular machines	1-20 nm	Assemblage of molecular components that are intended to carry out mechanical motions in response to external stimuli.
	Nanomotors	10nm-10μm	A nanoscale device that can transform energy from several sources such as chemical, optical, magnetic, ultrasonic, and electrical. Into self-propelled action
	DNA nanorobots	5-100nm	With the use of molecular recognition, DNA nanorobots can load and deliver medicament molecules to their targets.
Nanomanipulator	Nanorobotic manipulation based on	Macroscale	Tweezers are used as the robot end effector in tweezer-based

	optical/magnetic/acoustic tweezers		nanomanipulators, which handle nano-sized biological samples in a non-contact manner.
	Nanorobotic manipulation based on atomic force microscopy (AFM)	Macroscale	Nanomanipulators based on AFM use a nano-sized tip as the end effector to physically manipulate biological samples in a variety of environments, including air and liquids.
	Nanorobotic manipulation based on electron microscopy (EM)	Macroscale	Under the supervision of EM imaging, an EM-based nanomanipulator conducts robotic operations on biological samples in a vacuum or gaseous atmosphere.

Table 2

Biological samples of numerous sizes, ranging from single molecules in the human body, may be explored using a vast variety of nanorobotic devices. Ions, tiny molecules, and proteins may all be probed by molecular machines. Nano Manipulators based on tweezers can handle biological samples ranging in size from a few nanometers (like proteins) to tens of micrometers (such as cells). AFM/EM nano manipulators may manipulate biological materials at all levels, from single molecules to single cells to tissues. After injecting a large number of DNA nanorobots into a blood artery, the drug-carrying nanorobots can travel to sick spots in the body (such as tumors) and release medications to cure the diseased regions for in vivo applications.

The advent of DNA origami technology has opened up new possibilities for nanorobot research. DNA is a fundamentally programmable material, and by combining sequence-complementary domains, DNA molecules may be built into bespoke predesigned forms. Nanorobots can be made with DNA methods. DNA origami is a DNA-based device that folds a huge single strand of "scaffold" DNA into exact 2D and 3D structures stabilized by thousands of base pairs using programmed combinations of hundreds of short complementary "staple" oligonucleotides. Bottom-up self-assembly of discrete structures with sub nanometer fine features, dimensions ranging from the nanometer to the micrometer scale, and molecular weights up to the Giga Dalton scale is possible with DNA origami. A much wide range of quite functional static nanostructures & dynamic nanodevices have been produced using DNA origami (further specific information can be obtained in the literature), opening the path for single-molecule DNA nanorobotics.

The development of Atomic Force Microscopy i.e. AFM-based nanorobotic manipulation devices has opened up new possibilities for manipulating biological samples with extraordinary spatiotemporal resolution. AFM is a sophisticated technique that can explore native biological samples with nanoscale having spatial resolution & sub-100 millisecond temporal resolution under near-physiological settings (such as in liquids), making it useful in molecular and cell biology. The commercial AFM is primarily used for imaging and measuring. The nanorobotic manipulator, created by combining AFM with robotics, can handle nano-objects, bringing up many nouvelle possibilities for nanorobotics in biomedical applications.

CHALLENGES IN NON-INVASIVE MEDICAL DIAGNOSTICS

MICROELECTRONICS USED IN MEDICAL DIAGNOSTICS

Nowadays, micro-analytical devices are fitted with CMOS ASICs. The main benefit of using this is their miniaturization. In the few centimeters of space taken by the chip, massive systems can be fitted. Fluidics miniaturization and micromachined transducers allow this shrinking of electronics to be possible. In this field, there are two major categories of challenges which are needed to be resolved. First are challenges faced at circuit level, and second are packaging and interfaces challenges.

CARDIAC MAGNETIC RESONANCE AND COMPUTED TOMOGRAPHY ANGIOGRAPHY TO DIAGNOSE CAD

Both CMR and CCT allow a direct visual examination of the coronary architecture which improves their diagnostic accuracy and gives a predictive value which is negative. CMR is one of the newest imaging modalities for non-invasive CAD diagnosis. During stress tests, both direct analysis of coronary arteries and assessment of decrease in coronary reserve are used to accomplish this.

However, due to confounding variables such as the small size and convoluted character of the veins, as well as the continual heart and lung motions, magnetic resonance angiography utilized in coronary arteries cannot be visualized clearly and this remains a problem. CMR is not beneficial in detecting CAD in every case according to current guidelines. Currently, there is no data present which can help us analyze how CMR findings are useful and assess the risks in asymptomatic persons.

CHALLENGES IN EARLY-STAGE LUNG CANCER DETECTION

There is no clinical evaluation developed in aiding lung cancer at this time despite the new developments for cancer detection by the use of methylation biomarkers. ctDNA amount present in plasma in patients who have new malignancies(early-stage) is very low. This is a key restriction in using methods based on PCR for cancer detection in early stages. Smaller tumors are said to have less core necrosis, less vascularity, and less lymphatic invasion, all of which lead to less ctDNA fragments in the blood. (Aydin, 2007b)Researchers have found as few as 2,127 to 8,787 copies of cfDNA in 1 mL of plasma, which is comparable to 7–29 ng. The majority of cfDNA comes from blood leukocytes, and the amount of ctDNA found in plasma, in cancer patients who have smaller tumors, is 1 to 2 per mL of plasma. Because these methods have low specificity for diagnosing cancer in early stages, they are difficult to utilize for screening in healthy people. When samples undergo bisulfite conversion, almost 50 percent loss in sampling occurs, which lowers the test sensitivity. Furthermore, this method has a larger detection range than targeted deep sequencing techniques.

CHALLENGES IN DETECTION OF EXTRACRANIAL ARTERIAL DISEASE USING EEG

The electroencephalogram (EEG) is a non-invasive tool which is used for determining brain activity and state. EEG is used to monitor the brain and to diagnose a variety of neurological disorders. Current

advancements in techniques of ml and signal processing have made it possible to detect neurological events automatically in a variety of medical applications. The main issue in using EEG signals for real-time applications is to detect and predict seizures with high accuracy. When we apply these detection approaches to EEG signals using various methods, their resulting performance is found to be less than 80%. This value is much less than the expected performance value.

EEG has low signal amplitude (microvolts (V)) and is very sensitive to noise, which is its fundamental problem. When it comes to EEG signals, there are a number of drawbacks to be aware of. For starters, high amplitude EM fields and other factors tend to interfere with EEG recordings. Activation of muscles in the head region is the second constraint. Some of the other limitations are: (i) low ratio of signal-noise, and (ii) complex data analysis. Multiple analytics methods and various experiments are being studied to propose solutions to these problems. As a result, EEG is still one of the most direct and straightforward sources of data which is used in studying electrical brain activity phenomena.

CHALLENGES IN MANAGEMENT OF NON-SMALL CELL LUNG CANCER

The advancement in genetic heterogeneity shown among NSCLC patients has advanced dramatically, opening the door to tailored therapy for those with clinically identifiable genomic abnormalities. To make clinical decisions for patients with NSCLC, it is necessary to investigate the genetic abnormalities in the tumor. The gold standard which is used for tumor genotyping is tissue biopsy. However, the intricacy of molecular changes and difficulty with appropriate tumor tissue procurement are obstacles to tissue-based biomarker research. Furthermore, the geographic and temporal variability of genetic mutations within a patient might make it more difficult for testing and interpretation. If intra-patient heterogeneity of tumors is not represented effectively, then it limits efforts to grasp the complicated biology of malignancies. Furthermore, targeted therapy & chemotherapy that induce an adaptive effect in the tumor, & this is also vital from a therapeutic standpoint.

CONCLUSION

Wearable sweat biosensors are a form of wearable sensors that may be used to conduct physiological and clinical research in real-time. Despite tremendous advances by several organizations, some critical hurdles remain for sweat detection, such as on-body stability & the dependability of chemical sensors. The development of wearable power sources or the reduction of present wearable sweat sensor power consumption, on-demand programmable sweat biomarker extraction, and the link between sweat biomarker levels and particular physiological or behavioral outcomes are all urgently needed.

These difficulties, however, will be addressed by creative, collaborative, and multidisciplinary research.

Many studies employing wet-etching techniques to design La-doped-PZT (PLZT) & PZT films have recently been reported. To create a free-standing PZT force sensor with top and bottom electrodes creating a capacitor structure, a combination of wet-etch, ion beam etching (IBE), and deep reactive ion etching (DRIE) methods were used to create a unique wet etch recipe for patterning PZT films produced on Pt coated Si substrates. This sort of force sensor may be used to monitor sub-mN forces in minimally invasive surgical (MIS) equipment & palpable skin for robots, as exemplified by a micro-machined freestanding piezoelectric force sensor made of PZT thin film.

Using micromachining and wet etch methods, a freestanding PZT force sensor can be designed and constructed. A sol-gel method can be used to create PZT layers with a thickness of 1m. The microfabrication is done with a 5-mask method that includes IBE, DRIE,, and wet etch techniques. For patterning PZT, the wet-etch method offers good etch control, reduced undercut, retains the photoresist mask, and successfully eliminates residues on the etched surface. The SiO_2 and Si_3N_4 layers produced by PECVD serve as good masking layers for the DRIE and TMAH etch procedures, resulting in the successful release of a freestanding PZT force sensor (11.33 m thick) with dependable electrical properties. In terms of increased saturation polarization, the constructed force sensor has good ferroelectric characteristics.

In the era of individualized precision medicine, robotic nano manipulations based on nano manipulators provide a potential technique to diagnose and cure tumors at the nanoscale. Because drug response differs from person to person due to illness heterogeneity, precision medicine requires the capacity to predict & evaluate the efficacy of several therapies for a specific patient. Older & Traditional method of drug prediction approaches are mostly centered on detecting cellular biochemical qualities, even though pathogenic changes in cells are frequently accompanied by changes in cell physical properties. It's worth noting that there are still large gaps between present nanorobotics and the needs of biomedical communities, implying that there's still a lot of potentials for nanorobotics to improve for better biomedical applications. In vivo nanorobots now serve primarily as medication transporters, relying on specialized molecular recognition relationships. The motions of nanorobots are dependent on blood flow, and molecular-specific binding contacts are directly connected to the sometimes-adequate collision between particular molecules coated on nanorobots and target molecules on tumor cells. Also, it is now well recognized that detecting cell physics aids in understanding cell activity. Taken together, nanorobotic systems & their biomedical implementations along with applications necessitates fresh opportunities for linking robotics & biomedicine, which will have a substantial influence on the approaching era of biomedicine and artificial intelligence convergence. Nanorobotics for implementation in biomedical applications, in particular, is still in its premature or infancy stage, with plenty of space for progress. This will need collaboration among researchers and scientists from other departments & disciplines, and we have a lot to anticipate to as it develops.

REFERENCES

Augustine, R., Hasan, A., Primavera, R., Wilson, R. J., Thakor, A. S., & Kevadiya, B. D. (2020). Cellular uptake and retention of nanoparticles: Insights on particle properties and interaction with cellular components. *Materials Today Communications*, *25*, 101692. https://doi.org/10.1016/J.MTCOMM.2020.101692

Augustine, R., Mamun, A. al, Hasan, A., Salam, S. A., Chandrasekaran, R., Ahmed, R., & Thakor, A. S. (2021). Imaging cancer cells with nanostructures: Prospects of nanotechnology driven non-invasive cancer diagnosis. *Advances in Colloid and Interface Science*, *294*, 102457. https://doi.org/10.1016/J.CIS.2021.102457

Aydin, S. (2007a). A comparison of ghrelin, glucose, alpha-amylase and protein levels in saliva from diabetics. *Journal of Biochemistry and Molecular Biology*, *40*(1), 29–35. https://doi.org/10.5483/BMBREP.2007.40.1.029

Aydin, S. (2007b). A comparison of ghrelin, glucose, alpha-amylase and protein levels in saliva from diabetics. *Journal of Biochemistry and Molecular Biology*, *40*(1), 29–35. https://doi.org/10.5483/BMBREP.2007.40.1.029

Bandodkar, A. J., Molinnus, D., Mirza, O., Guinovart, T., Windmiller, J. R., Valdés-Ramírez, G., Andrade, F. J., Schöning, M. J., & Wang, J. (2014). Epidermal tattoo potentiometric sodium sensors with wireless signal transduction for continuous non-invasive sweat monitoring. *Biosensors & Bioelectronics*, *54*, 603–609. https://doi.org/10.1016/J.BIOS.2013.11.039

Bandodkar, A. J., & Wang, J. (2014). Non-invasive wearable electrochemical sensors: a review. *Trends in Biotechnology*, *32*(7), 363–371. https://doi.org/10.1016/J.TIBTECH.2014.04.005

Chaki, N. K., & Vijayamohanan, K. (2002). Self-assembled monolayers as a tunable platform for biosensor applications. *Biosensors & Bioelectronics*, *17*(1–2), 1–12. https://doi.org/10.1016/S0956-5663(01)00277-9

Chandrasekaran, R., Madheswaran, T., Tharmalingam, N., Bose, R. J., Park, H., & Ha, D. H. (2021). Labeling and tracking cells with gold nanoparticles. *Drug Discovery Today*, *26*(1), 94–105. https://doi.org/10.1016/J.DRUDIS.2020.10.020

Chung, M., Fortunato, G., & Radacsi, N. (2019). Wearable flexible sweat sensors for healthcare monitoring: a review. *Journal of the Royal Society, Interface*, *16*(159). https://doi.org/10.1098/RSIF.2019.0217

Corrie, S. R., Coffey, J. W., Islam, J., Markey, K. A., & Kendall, M. A. F. (2015). Blood, sweat, and tears: developing clinically relevant protein biosensors for integrated body fluid analysis. *The Analyst*, *140*(13), 4350–4364. https://doi.org/10.1039/C5AN00464K

Dorn, L. D., Lucke, J. F., Loucks, T. L., & Berga, S. L. (2007). Salivary cortisol reflects serum cortisol: analysis of circadian profiles. *Annals of Clinical Biochemistry*, *44*(Pt 3), 281–284. https://doi.org/10.1258/000456307780480954

Falk, M., Psotta, C., Cirovic, S., & Shleev, S. (n.d.). *Non-Invasive Electrochemical Biosensors Operating in Human Physiological Fluids*. https://doi.org/10.3390/s20216352

Falk, M., Psotta, C., Cirovic, S., & Shleev, S. (2020). Non-Invasive Electrochemical Biosensors Operating in Human Physiological Fluids. *Sensors (Basel, Switzerland)*, *20*(21), 1–28. https://doi.org/10.3390/S20216352

Fye, W. B. (1994). A history of the origin, evolution, and impact of electrocardiography. *The American Journal of Cardiology*, *73*(13), 937–949. https://doi.org/10.1016/0002-9149(94)90135-X

Green-Church, K. B., Nichols, K. K., Kleinholz, N. M., Zhang, L., & Nichols, J. J. (2008). Investigation of the human tear film proteome using multiple proteomic approaches. *Molecular Vision*, *14*, 456. /pmc/articles/PMC2268847/

Guilbault, G. G., Palleschi, G., & Lubrano, G. (1995). Non-invasive biosensors in clinical analysis. *Biosensors & Bioelectronics*, *10*(3–4), 379–392. https://doi.org/10.1016/0956-5663(95)96856-T

Ilea, A., Andrei, V., Feurdean, C. N., Băbtan, A. M., Petrescu, N. B., Câmpian, R. S., Bosca, A. B., Ciui, B., Tertis, M., Săndulescu, R., & Cristea, C. (2019). Saliva, a Magic Biofluid Available for Multilevel Assessment and a Mirror of General Health-A Systematic Review. *Biosensors*, *9*(1). https://doi.org/10.3390/BIOS9010027

Kanoun, O., & Tränkler, H. R. (2004). Sensor technology advances and future trends. *IEEE Transactions on Instrumentation and Measurement*, *53*(6), 1497–1501. https://doi.org/10.1109/TIM.2004.834613

Karpova, E. v., Karyakina, E. E., & Karyakin, A. A. (2020). Wearable non-invasive monitors of diabetes and hypoxia through continuous analysis of sweat. *Talanta*, *215*. https://doi.org/10.1016/J.TALANTA.2020.120922

Kim, J., Campbell, A. S., de Ávila, B. E. F., & Wang, J. (2019). Wearable biosensors for healthcare monitoring. *Nature Biotechnology*, *37*(4), 389. https://doi.org/10.1038/S41587-019-0045-Y

Larpant, N., Pham, A. D., Shafaat, A., Gonzalez-Martinez, J. F., Sotres, J., Sjöholm, J., Laiwattanapaisal, W., Faridbod, F., Ganjali, M. R., Arnebrant, T., & Ruzgas, T. (2019). Sensing by wireless reading Ag/AgCl redox conversion on RFID tag: universal, battery-less biosensor design. *Scientific Reports*, *9*(1). https://doi.org/10.1038/S41598-019-49245-3

Li, S., Sack, R., Vijmasi, T., Sathe, S., Beaton, A., Quigley, D., Gallup, M., & Mcnamara, N. A. (2008). Antibody Protein Array Analysis of the Tear Film Cytokines. *Optometry and Vision Science : Official Publication of the American Academy of Optometry*, *85*(8), 653. https://doi.org/10.1097/OPX.0B013E3181824E20

Liao, J. C., Mastali, M., Gau, V., Suchard, M. A., Møller, A. K., Bruckner, D. A., Babbitt, J. T., Li, Y., Gornbein, J., Landaw, E. M., McCabe, E. R. B., Churchill, B. M., & Haake, D. A. (2006). Use of electrochemical DNA biosensors for rapid molecular identification of uropathogens in clinical urine specimens. *Journal of Clinical Microbiology*, *44*(2), 561–570. https://doi.org/10.1128/JCM.44.2.561-570.2006

Liu, J., Geng, Z., Fan, Z., Liu, J., & Chen, H. (2019). Point-of-care testing based on smartphone: The current state-of-the-art (2017-2018). *Biosensors & Bioelectronics*, *132*, 17–37. https://doi.org/10.1016/J.BIOS.2019.01.068

Mach, K. E., Wong, P. K., & Liao, J. C. (2011). Biosensor diagnosis of urinary tract infections: a path to better treatment? *Trends in Pharmacological Sciences*, *32*(6), 330–336. https://doi.org/10.1016/J.TIPS.2011.03.001

Malon, R. S. P., Sadir, S., Balakrishnan, M., & Córcoles, E. P. (2014). Saliva-Based Biosensors: Noninvasive Monitoring Tool for Clinical Diagnostics. *BioMed Research International*, *2014*. https://doi.org/10.1155/2014/962903

Markoulli, M., Papas, E., Petznick, A., & Holden, B. (2011). Validation of the flush method as an alternative to basal or reflex tear collection. *Current Eye Research*, *36*(3), 198–207. https://doi.org/10.3109/02713683.2010.542867

Mitchell, J. S., Lowe, T. E., & Ingram, J. R. (2009). Rapid ultrasensitive measurement of salivary cortisol using nano-linker chemistry coupled with surface plasmon resonance detection. *The Analyst*, *134*(2), 380–386. https://doi.org/10.1039/B817083P

Morris, L. R., McGee, J. A., & Kitabchi, A. E. (1981). Correlation between plasma and urine glucose in diabetes. *Annals of Internal Medicine*, *94*(4 pt 1), 469–471. https://doi.org/10.7326/0003-4819-94-4-469

Moyer, J., Wilson, D., Finkelshtein, I., Wong, B., & Potts, R. (2012). Correlation between sweat glucose and blood glucose in subjects with diabetes. *Diabetes Technology & Therapeutics*, *14*(5), 398–402. https://doi.org/10.1089/DIA.2011.0262

Munje, R. D., Muthukumar, S., Panneer Selvam, A., & Prasad, S. (2015). Flexible nanoporous tunable electrical double layer biosensors for sweat diagnostics. *Scientific Reports*, *5*. https://doi.org/10.1038/SREP14586

Nyein, H. Y. Y., Gao, W., Shahpar, Z., Emaminejad, S., Challa, S., Chen, K., Fahad, H. M., Tai, L. C., Ota, H., Davis, R. W., & Javey, A. (2016). A Wearable Electrochemical Platform for Noninvasive Simultaneous Monitoring of Ca(2+) and pH. *ACS Nano*, *10*(7), 7216–7224. https://doi.org/10.1021/ACSNANO.6B04005

Olarte, O., Chilo, J., Pelegri-Sebastia, J., Barbe, K., & van Moer, W. (2013). Glucose detection in human sweat using an electronic nose. *Annual International Conference of the IEEE Engineering in Medicine and Biology Society. IEEE Engineering in Medicine and Biology Society. Annual International Conference*, *2013*, 1462–1465. https://doi.org/10.1109/EMBC.2013.6609787

Pan, Y., Sonn, G. A., Sin, M. L. Y., Mach, K. E., Shih, M. C., Gau, V., Wong, P. K., & Liao, J. C. (2010). Electrochemical immunosensor detection of urinary lactoferrin in clinical samples for urinary tract infection diagnosis. *Biosensors & Bioelectronics*, *26*(2), 649–654. https://doi.org/10.1016/J.BIOS.2010.07.002

Pappa, A. M., Curto, V. F., Braendlein, M., Strakosas, X., Donahue, M. J., Fiocchi, M., Malliaras, G. G., & Owens, R. M. (2016). Organic Transistor Arrays Integrated with Finger-Powered Microfluidics for Multianalyte Saliva Testing. *Advanced Healthcare Materials*, *5*(17), 2295–2302. https://doi.org/10.1002/ADHM.201600494

Park, K. (2007). Nanotechnology: What it can do for drug delivery. *Journal of Controlled Release*, *120*(1–2), 1–3. https://doi.org/10.1016/J.JCONREL.2007.05.003

Raffaelli, M., de Crea, C., & Bellantone, R. (2019). Laparoscopic adrenalectomy. *Gland Surgery*, *8*, S41–S52. https://doi.org/10.21037/GS.2019.06.07

Rocchitta, G., Spanu, A., Babudieri, S., Latte, G., Madeddu, G., Galleri, G., Nuvoli, S., Bagella, P., Demartis, M. I., Fiore, V., Manetti, R., & Serra, P. A. (2016). Enzyme Biosensors for Biomedical Applications: Strategies for Safeguarding Analytical Performances in Biological Fluids. *Sensors (Basel, Switzerland)*, *16*(6). https://doi.org/10.3390/S16060780

Sack, R., Conradi, L., Beaton, A., Sathe, S., McNamara, N., & Leonardi, A. (2007). Antibody array characterization of inflammatory mediators in allergic and normal tears in the open and closed eye

environments. *Experimental Eye Research*, *85*(4), 528–538. https://doi.org/10.1016/J.EXER.2007.07.004

Segal, A., & Wong, D. T. (2008). Salivary diagnostics: Enhancing disease detection and making medicine better. *European Journal of Dental Education*, *12*(SUPPL. 1), 22–29. https://doi.org/10.1111/j.1600-0579.2007.00477.x

Siddaiah-Subramanya, M., Tiang, K. W., & Nyandowe, M. (2017). A New Era of Minimally Invasive Surgery: Progress and Development of Major Technical Innovations in General Surgery Over the Last Decade. *The Surgery Journal*, *3*(4), e163. https://doi.org/10.1055/S-0037-1608651

Spehar-Délèze, A. M., Anastasova, S., & Vadgama, P. (2021). Monitoring of Lactate in Interstitial Fluid, Saliva and Sweat by Electrochemical Biosensor: The Uncertainties of Biological Interpretation. *Chemosensors 2021, Vol. 9, Page 195*, *9*(8), 195. https://doi.org/10.3390/CHEMOSENSORS9080195

Trigona, C., Graziani, S., & Baglio, S. (2020a). Changes in sensors technologies during the last ten years: Evolution or revolution? *IEEE Instrumentation and Measurement Magazine*, *23*(6), 18–22. https://doi.org/10.1109/MIM.2020.9200876

Trigona, C., Graziani, S., & Baglio, S. (2020b). Changes in sensors technologies during the last ten years: Evolution or revolution? *IEEE Instrumentation and Measurement Magazine*, *23*(6), 18–22. https://doi.org/10.1109/MIM.2020.9200876

Wilson, R., & Turner, A. P. F. (1992). Glucose oxidase: an ideal enzyme. *Biosensors and Bioelectronics*, *7*(3), 165–185. https://doi.org/10.1016/0956-5663(92)87013-F