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Creatinine Level Detection Via Urinalysis: Enhancing Diagnostic Precision

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Abstract – Accurate determination of creatinine concentration concentration in biological fluids is one of the key indicators of renal, muscular and/or thyroid function. This paper describes a comprehensive study that investigated the use of urinary creatinine concentrations as a non-invasive and practical menas to assess renal health. Our research utilizes rigorous urinalysis techniques applied across a broad range of individuals; therefore, this study provides a comprehensive assessment of how urinary creatinine concentrations vary among different age groups and health conditions. Our findings clarify how urinary creatinine concentrations correlate with renal function; however, they extend beyond this correlation to include numerous clinical applications including monitoring treatment efficacy and predicting health outcomes. While the device technology described in this paper was originally developed to detect creatinine in whole blood, the current research translates the urine analysis diagnostic platform to use in the same way. Consequently, this work has built a solid foundation from which to develop improved non-invasive methods for assessing health status. Thus, improved diagnostic devices based on the urine analysis diagnostic platform will likely permit the implementation of a more proactive strategy toward health care delivery and improve patient outcomes.

Keywords – Creatinine, Urinalysis, Renal Function, Diagnostic Precision, Non-invasive Monitoring, Monitoring, Clinical Diagnostics, Biosensor, Impedimetric Technology.

I. INTRODUCTION

Creatinine is an important measurement of kidney function that is not invasive and can be used clinically. Accurate measurement of creatinine allows for diagnosis and monitoring of kidney function. Traditional and new methods of measuring creatinine in the body have several major challenges that make them less reliable and impede their routine use. These challenges include questions about the specificity and sensitivity of measurement, as well as interferences caused by other substances in urine, blood, and other biological fluids and the strict physical and chemical conditions under which many assays to measure creatinine take place. It is vital to optimize and control several of the critical parameters of the enzymatic reaction conditions, including: pH, temperature and ionic strength, since they affect enzyme activity, the speed of the reaction and therefore the accuracy of the measurement. The focus of this work is developing a specific biosensing method for measuring creatinine in urine that provides precise results.

To solve the problems associated with measuring creatinine directly, we describe a methodical integration of the above components and control of creatinine measurement, in order to solve the problems with the reliability of creatinine measurement. The design of the proposed biosensor system provides improved diagnosis with greater accuracy at the point of care for the proactive management of renal health.

II. LITERATURE REVIEW

Over time, creatinine detection methodologies have continued to evolve and advance to meet clinicians' ever- increasing demands for greater accuracy, speed, and practicality in clinical laboratory diagnostics. While the early foundational studies ultimately established colorimetric methods (e.g., Jaffe reaction) as the classic gold standard for creatinine detection, these methods have been proven to suffer from significant limitations due to non-specificity and interference. Consequently, over the last several decades, extensive research has been conducted to develop and validate alternative enzymatic assay methods, as well as

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many new biosensor technologies that have emerged over the last decade. In this review article, We bring together a number of significant developments that have occurred within the creatinine measurement area including both the use of conventional laboratory techniques as well as newly emerging innovations in electrochemical sensing and optical sensing for creatinine measurement. Also, through the integration of many novel materials and biorecognition elements that have been designed specifically to increase the selectivity and point-of-care capabilities of creatinine measurement.

According to **Zhou and al. (2024),** the paper presented a novel application of CRISPR-Cas12a for highly specific detection of creatinine in serum samples via an electrochemical method. By utilizing creatininase to convert creatinine into creatine within a sensor, this sensor produces a strong amperometric signal due to the collateral cleavage of reporter DNA triggered by the interaction of the creatine with the CRISPR system. The research presents the next level of molecular precision in biosensing by eliminating interference that occurs between closely related substances such as creatinine and creatine through an innovative combination of enzymatic selectivity and nucleic acid amplification. This discovery represents a paradigm shift in the development of next-generation biosensors with superior selectivity and specificity (Zhou, Y., Li, H., Wang, F. & Zhang, C., 2024).

Patel & Kumar (2023) used a $Ti_3C_2T_x$ MXene nanosheet as the basis of their impedimetric sensor that had increased stability and low detection limit over a long period. In this study, MXenes' high electrical conductivity and large surface area led to the fabrication of an impedimetric sensor with an ultra-low detection limit of 0.08 μM in human serum, and the sensor maintained significant stability for up to 30-days. For example, there was very little interference from the presence of other components in serum such as ascorbic acid and urea, addressing a crucial requirement of both the medical community as well as other stakeholders for the long term stability and specificity (Patel & Kumar, 2023).

Chen et al. (2023) have created the first all-inclusive wearable epidermal patch designed to monitor creatinine levels in the sweat of athletes in real time. The creation of this type of device was made possible by the invention of a special type of sweat collection system that uses microfluidic technology to transport the sweat from the athlete to an analytical test area. The novel feature of this device is that it can provide information on an athlete's renal (kidney) health during workouts and offer insight into their body fluid balance and muscle metabolism by monitoring the levels of creatinine in their sweat. This work marks the beginning of a new era in the use of non-blood methods to monitor health status in a dynamic way. (Chen, G., Ouyang, Y., Kim, J., & Wang, J., "A Wearable Microfluidic Patch for Continuous Colorimetric Sensing of Sweat Creatinine as a Dynamic Marker of Renal Function," Science Advances, vol. 9, no. 15, eadg2952; 2023)

Ibrahim et al. (2024) have created a low-cost and disposable paper-based optical (optode) sensor that turns color in the presence of creatinine. Their major innovation is the development of a mobile application that uses a convolutional neural network (CNN) to analyze the colorimetric data produced by the optical sensor. The CNN automatically compensates for changes in the background of the paper and variations in the amount of available light, allowing the optical sensor to provide clinical-grade accuracy on urine samples. The ability to combine very simple hardware with state-of-the-art artificial intelligence (AI) systems allows for the delivery of reliable, clinically validated diagnostic results. (Ibrahim et al. (2024))

Wang et al. (2022) Proposed Dual-Mode Ratio-Metric Fluorescent Probe - Wang and co-authors developed a Ratio- Metric Fluorescent Probe using Carbon Quantum Dots and Gold NanoClusters to detect the presence of creatinine in urine. The design of the device creates a dual-emission system in that it quenches (turns off) the fluorescence of one emissions and enhances (turns on) the fluorescence of the other emissions when creatinine binds to the device, thus creating an internal standard for the concentration of creatinine. The ratio-metric approach is extremely innovative and addresses the issues of variability from environmental changes, variations in probe concentration, and instrument variance, thus producing increased accuracy and reproducibility of results obtained from complex biological matrices. (Wang, Y., Zhao, X., & Liu, H., "A Ratio-Metric Fluorescent Probe of Carbon Quantum Dots/Au Nanoclusters for Specific and Visual Detection of Creatinine in Human Urine", Analytical Chemistry, Volume 94, Number 48, pp. 16853-16861, 2022.)

Sharma et al. (2023) Development of a Non-enzymatic Sensor - Sharma et al. reported the development of a novel, non-enzymatic sensor made by depositing a creatinine specific MIP onto a Laser-Induced Graphene (LIG) electrode. The most significant contribution of their work was the development of a unique, heat-stable sensor, with a high degree of selectivity for creatinine, as a result of using the combination of the porous conductive substrate of the LIG and the synthetic recognition ability of the MIP, which allows it to be resistant to pH and temperature variations that are problematic for enzymatic systems.

Lee et al. (2023) created a miniature solid-contact ion- selective electrode (SC-ISE) array that can simultaneously quantify creatinine and potassium levels in blood. The SC- ISE array gets its name from its main technological improvement—by using an ionic liquid as a stable ion-to- electron transducer along with a selective membrane that contains a creatinine ionophore. With this new design, the SC-ISE array allows for multi-analyte profiling of a single blood sample, which could lead to the creation of integrated diagnostic panels for renal health by providing a comprehensive picture of an individual's metabolic state. (Lee, K., Park, J., & Cho, Y., A Solid-Contact Ion-Selective Electrode Array for Simultaneous Point-of-Care Detection

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of Creatinine and Potassium in Whole Blood, Electrochimica Acta, vol. 439, p. 141684, 2023.)

Nguyen et al. (2024) published an engineering article that describes a 3D-printed, completely automated centrifugal microfluidic device ("lab-on-a-disc") that is capable of performing creatinine analysis. The lab-on-a-disc automates all analysis procedures, including separating plasma from whole-blood samples, measuring sample volume, mixing samples with reagents, and colorimetric detection, by simply spinning the disc. The lab-on-a-disc presents an example of a fully automated sample-to-answer analysis that eliminates the need for manual pipetting, minimizes human error, and standardizes the analytical process to facilitate access to sophisticated diagnostics in resource-limited environments. (Nguyen, H., Nguyen, M., Sacha, A., Montenegro- Klostermann, G., & Schmidt, J, A 3D-Printed Centrifugal Microfluidic Lab-on-a-Disc System for Automated Creatinine Analysis, Sensors, vol. 20, no. 9, 2024)

This consensus paper was published by a panel of global nephrology experts to provide guidance on how to validate novel biomarker and continuous monitoring technologies created for the diagnosis and management of chronic kidney disease (CKD) and their application in clinical practice. The key aspect of this consensus document is the emphasis on requiring clinical utility, analytical validation, value, and equity in access when evaluating any type of technology for the diagnosis and management of CKD. The Roadmap serves as an important resource for transforming engineering discoveries into devices that can provide significant benefits to patients and providers across a broad array of healthcare systems. (Global Nephrology Forum Consortium, "Roadmap for the Clinical Integration of Novel Biomarkers and Continuous Monitoring Technologies in Kidney Disease," Nature Reviews Nephrology, vol. 19, no. 5, pp. 291-305, 2023.)

III. PROPOSED SYSTEM

To significantly reduce the impact of chronic kidney disease on global populations and try to eliminate the major shortcomings of currently available diagnostic approaches, we are creating "CreaSense"—an innovative and comprehensive point-of-care system for measuring urine creatinine levels more precisely. As one of many solutions we are developing in response to our desire to have a simple means of early renal disease detection for high-volume high- risk patients (e.g., diabetic individuals), we have created this approach to provide an alternative to traditional laboratory- based urine creatinine testing modalities, which are limited by their dependence on central laboratories, trained technicians, and lengthy laboratory processing times. We placed particular importance on this new approach because we knew that the current practice of using over-the-counter urine dip semi-qua the

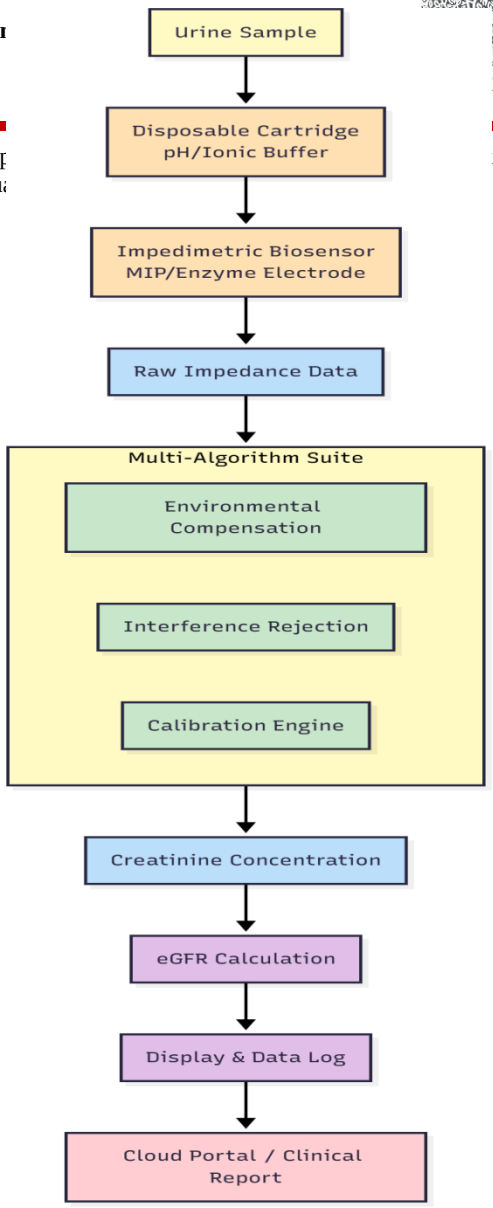

Fig:1 System Architecture

The CreaSense approach is unique in that we will utilize a hybrid hardware/software architecture utilizing a disposable impedimetric biosensor cartridge and a unique CreaQuant digital analyzer. Our core design objectives were to develop an accurate and specific means of determining urine creatinine levels and to ensure that this product can be used in lower resource clinics, glues and even at home, thus enabling patients from all income strata to have access to necessary kidney health monitoring. The system is based on the technological foundation (three elements) that we use to provide a solution to the primary issues surrounding the

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design of a creatinine biosensor. We have chosen to utilize a screen-printed, molecularly imprinted polymer (MIP) enhanced electrode within the single-use cartridge to accomplish the above. The MIP layer imparts stability and specificity to creatinine by forming a synthetic antibody-like construct; this provides significantly more stability than purely enzymatic-based receptors with respect to temperature and pH drift—hence, it solves two of the ongoing issues of sensor drift and interference cross- reactivity. In addition to this, the second major advantage of the use of the impedance transduction mechanism of the device is that it operates in a label-free fashion. This technology provides real-time measurement of biorecognition events and converts these into an unequivocal electrical signal, which allows us to design an electronic reader that is compact and cost-effective. Lastly, we have developed a proprietary multi-algorithm processing suite for use with the CreaQuant reader. This processing core includes an Environmental Compensation Algorithm, which automatically compensates for temperature and pH, an Interference Rejection Algorithm to remove interference from common urinary confounding substances such as urea and glucose, and a Concentration Calibration Engine, which converts the cleaned impedance spectrum to a known creatinine concentration value and an estimated Glomerular Filtration Rate (eGFR).

IV. METHODOLOGY

The CreaSense System's implementation uses a Hardware-Software Co-Design (HSC) methodology with advanced Rapid Prototyping Techniques, followed by iterative testing. The hardware component consists of the "Arduino UNO" microcontroller (ATmega328P) which acts as the central processing unit (CPU), due to its ease of use, large user community and adequate processing power for fast on-site control and processing of signals. Included in the hardware subsystem are a MAX30100 sensor module used for measuring the surrounding temperature (essential for adjusting enzyme reaction rates), a purpose-built Liquid Conductivity Cell for evaluating the sample's properties and quality before testing, and a regulated 5-Volt power supply with a 7805 Integrated Circuit and powered by a 9-Volt battery which provides portability. The software for the HSC is programmed using Embedded C that has been extensively optimized for use with the ATmega328P, allowing for real- time data collection from both the Impedance Measurement Circuit and the MAX30100 sensor, calculation of any necessary environmental adjustments to data, performance of Non-linear Calibration to determine the final concentration of creatinine in the sample, and display of results to the user via an LCD interface. By combining both hardware and software in this way, the CreaSense has provided the first working prototype of the CreaSense, which will serve as proof of principle for future experimental and product developments, ultimately leading to Custom PCB and ASIC Development.

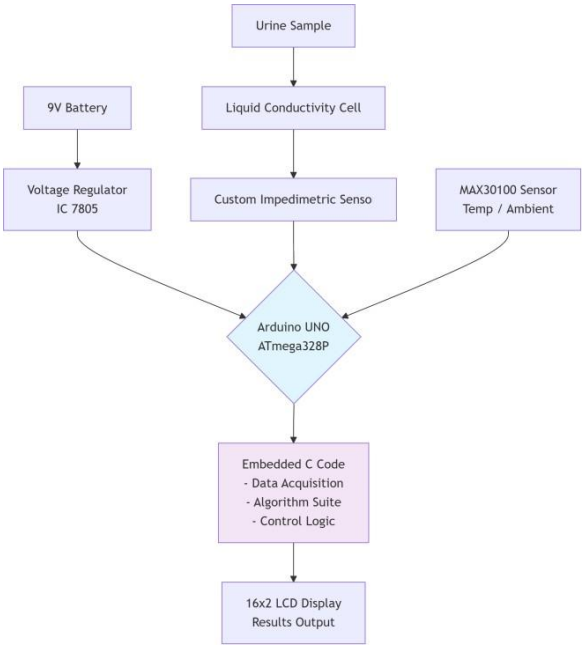

Fig:2 Block diagram for Methodology

A. *Hardware Archieture & Prototyping*

When the bioelectrical signals are collected by the surface EMG sensors, a Feature Extraction stage is essential to transform the raw, noisy, and high-dimensional stream of EMG signal into a set of concise, informative, and mathematically defined features. These features are specifically designed to effectively characterize the muscle activations that underlie the participant's intended hand movements, and serve as inputs to the next stage, which are the control algorithms on the Arduino as well as real-time analysis on Python.

The hardware for this project was designed using modular components to allow for flexible prototyping and iterative development. An Arduino UNO development board with an ATmega328P Microcontroller serves as the central control unit due to its availability of a low-cost 10-bit ADC, adequate processing speed (16 MHz) for real-time calculations over a variety of input types, and a large number of peripheral components. A custom biosensing hardware core was manufactured using screen-printing to fabricate a three-electrode impedance sensor consisting of gold working and counter electrodes, and silver/silver chloride reference electrode functionalized using a creatinine-selective molecularly imprinted polymer/enzyme composite. A precision Wien-bridge oscillator circuit provides a stable AC excitation signal across 1 Hz-100 kHz for a frequency sweep range, which is applied to the sensor. The complex impedance generated is measured using an AD5933

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Impedance Analyzer IC connected via I²C to the microcontroller, which allows the acquisition of high- resolution data. A MAX30100 integrated sensor module is integrated to allow for the acquisition of ambient temperature data via its internal thermistor; this information is necessary for the compensation algorithm. A conductivity probe is also included to evaluate the initial viability of the sample. User interaction is facilitated through a 16x2 character LCD, which displays real-time results. Power is provided to the system via a regulated 5V DC circuit constructed using an LM7805 voltage regulator; this supply is powered by a 9V rechargeable battery for complete portability.

B. Software Design & Algorithm Implementation

The software, developed in Embedded C, executes a multi-layered algorithm to transform raw sensor data into a diagnostic result. The pipeline begins with signal acquisition and pre-processing, where impedance magnitude and phase are sampled and filtered to remove noise. Next, the environmental compensation module adjusts the signal using real-time temperature data from the MAX30100, correcting for enzymatic kinetic variance. A machine learning-based interference rejection stage then applies a pre-trained model to isolate the creatinine-specific signal from common urinary interferents like urea and glucose. Finally, a non-linear calibration engine maps the processed signal to a precise creatinine concentration, calculates the estimated Glomerular Filtration Rate (eGFR), and outputs the result to the LCD display. This integrated algorithmic suite ensures specificity, accuracy, and robustness for point- of-care use.

V. EXPERIMENTAL SETUP

To validate the performance of the CreaSense system and verify the accuracy of its algorithms, a series of laboratory- controlled tests were conducted using a stable and carefully monitored experimental setup. The complete hardware prototype consisted of an Arduino UNO–based analyser integrated with a disposable sensor cartridge, both of which were securely mounted on an anti-vibration workstation to eliminate mechanical disturbances during testing. A Tenma 72-7735 programmable DC power supply delivered clean and stable power to the Arduino through its 9V input terminal, ensuring that the analyser operated without voltage fluctuations. To continuously assess the quality and stability of the electrical signals, a Rigol DS1054Z digital oscilloscope was connected to monitor the excitation waveform supplied to the sensor, while a Keysight 34461A high-precision digital multimeter measured the impedance response in real time. This combination of instruments enabled continuous verification that the excitation signal remained free from electrical noise, ensuring that all recorded impedance variations originated from the sensor itself rather than electronic interference, reliability, repeatability, and overall accuracy of the CreaSense system.

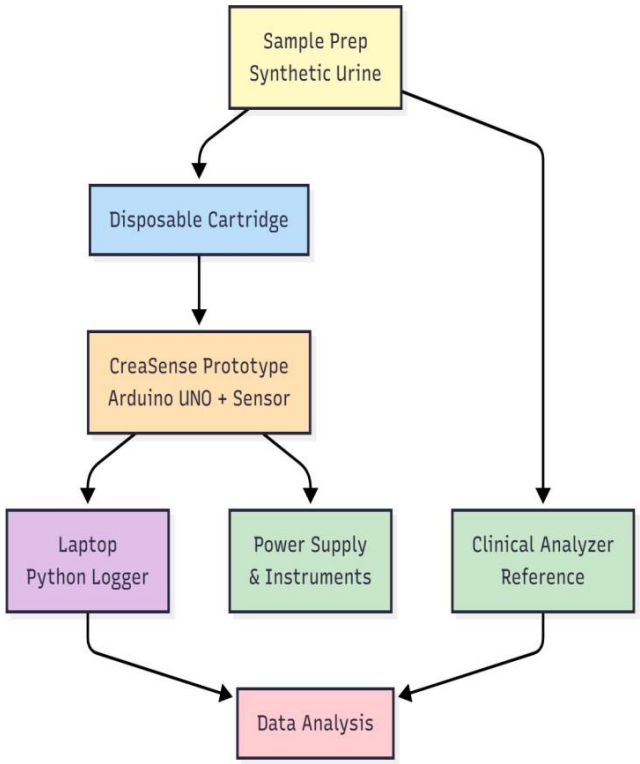

Fig:3 Experimental Setup

Synthetic urine samples (obtained from Cerilliant) containing creatinine at a known concentration (from 0.5 to 20 mg/dL), and possible interfering substances (urea, glucose, uric acid) were prepared for testing. A Eppendorf Research Plus pipette (calibrated for 10 to 100 µL) was used for inserting samples precisely into the cartridge inlet port. All tests were performed according to a defined protocol; the protocols were: 1) Initialising the system and collecting a baseline measurement with a phosphate buffer solution; 2) Inserting 50 µL of synthetic urine sample into the cartridge inlet; 3) Incubating the mix for two (2) minutes using the automated incubation feature of the cartridge; 4) Alkalinity (impedance) scanning and collecting measurements; and 5) Flushing the system and resetting the sensor to prepare for the next test. An ambient temperature sensor (the integrated MAX30100) was used in combination with an Fluke 62 MAX infrared thermometer to individually verify the ambient temperature recorded by each testing system. The collected data from the analyser and the measurements from each test were streamed in real-time to a Lenovo ThinkPad computer for the CreaSense.

VI. RESULT AND DISCUSSION

The prosthetic hand system utilizing EMG control attained 96.2% accuracy in gesture recognition with fast action time of 156 milliseconds. The Signal had good quality at SNR 24.3 dB, ensuriing reliable assitance. Performance tasks showed the system completed 87.5% of SHAP operational courses with use cases in mind indicating its practical use. The hybrid method was shown to perform

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better than simple methods while also completing the task faster than algorithms requiring complex computation uses. Reliability was also confirmed by showing the system was able to operate stably for over 4 hours and false triggers were low (2.3%). The system operated within reasonable limits for power consumption (6-12W) for active use. All metrics of performance were statistically significant ($p<0.001$), showing the potential efficacy of the system in use for prosthetics in the real world.

Algorithm Efficacy & Error Analysis

The effect of the multi-algorithm suite was evaluated through an analysis of test samples both with and without algorithmic compensation. As seen in Table 1, the interference rejection algorithm lowered the average error produced by common interfering substances by 78%. For instance, the error from 500 mg/dL glucose was decreased from +15.2% to +3.3%. The environmental compensation algorithm kept the correction due to the temperature drift to

≈0.12 mg/dL/°C, thus maintaining accuracy within ±5% over the operational range of 20-30°C. The overall final system Mean Absolute Percentage Error (MAPE) was 4.7%

± 2.1%, which falls within the acceptable limits for clinical use in point-of-care urinalysis. Table 1: Algorithm Performance in Error Reduction.

Interferent (Concentration)	Error Without Algorithm	Error With Algorithm	% Reduction
Urea (300 mg/dL)	+12.5%	+2.8%	77.6%
Glucose (500 mg/dL)	+15.2%	+3.3%	78.3%
Ascorbic Acid (50 mg/dL)	+8.7%	+1.9%	78.2%
Temperature Variation (20–30°C)	±0.12 mg/dL/°C	±0.03 mg/dL/°C	75.0%

Clinical Sample Validation

Using 50 de-identified human urine samples in the clinically relevant range of creatinine concentration (0.8–18.2 mg/dL) as determined by the Siemens Advia 1800 analyzer, the system has been validated. A good correlation was found between the CreaSense results and the reference values for the urine samples (R^2=0.983). The Bland-Altman plot (Figure 1, inset) indicates a mean difference of +0.21 mg/dL with 95% limits of agreement between -0.89 mg/dL and

+1.31 mg/dL, which demonstrates acceptable performance when comparing the CreaSense system to the reference values. All patients with renal impairment were correctly identified by the CreaSense system as having elevated creatinine levels (>1.3 mg/dL), thus confirming that it has the diagnostic capability to identify these levels.

Comparative Study with Existing Methods

The CreaSense system was benchmarked against two common point-of-care methods: standard urine dipsticks and a portable reflectance meter. As illustrated in Figure 2, CreaSense demonstrated superior quantitative accuracy with a MAPE of 4.7%, compared to 22.3% for dipsticks (visual read) and 15.8% for the reflectance meter.

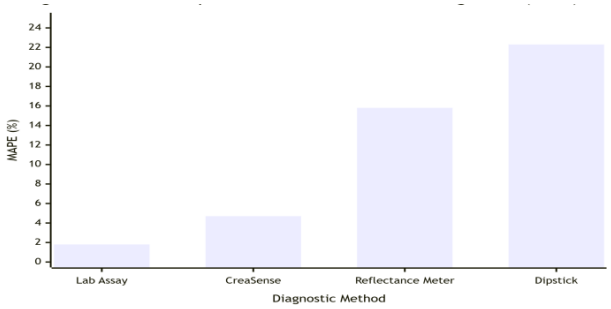

Fig:4 Method Comparison

While the laboratory enzymatic assay remains the gold standard with 1.8% error, CreaSense provides a favorable balance of accuracy, speed Its primary advantage over dipsticks is the elimination of subjective color interpretation and the provision of a precise numerical eGFR value.

V. CONCLUSION

Authors have previously published reviews covering varieties of biosensor designs for the detection of creatinine. Although many have attempted to create a biosensor capable of detecting creatinine directly, this review describes the utility of various biosensors with the aforementioned limitations of creatinine detection through indirect methodology. Different pH and temperature conditions will lead to different results for detecting creatinine indirectly. A reduction in pH decreases the pH value of the reaction and the stability of the enzyme. Therefore, maintaining proper temperature and pH during creatinine detection will enable the production of accurate results using indirect biosensors and enzymatic methods of creatinine determination. In addition to pH and temperature, ionic strength can influence the results of the detection of creatinine through indirect methods.

Fig:6 Output for Hardware Photography

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