cirCAT: PURRtentio: a Litter Box that Monitors Feline Urine using Electrochemical Biosensors

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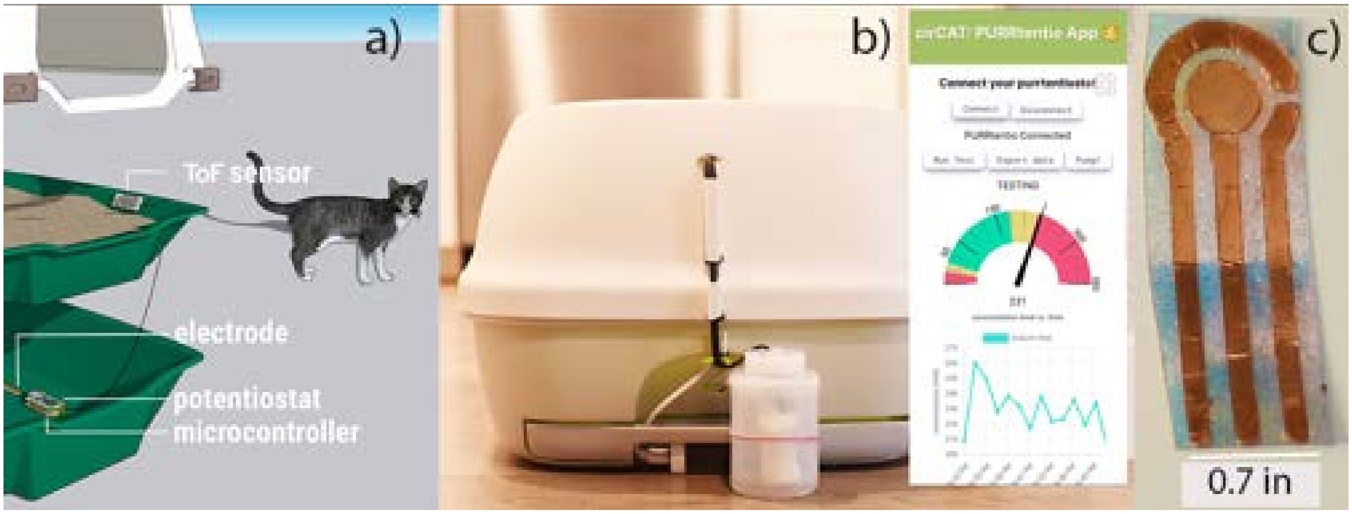

Figure 1: PURRtentio a) System components. b) Litter Box System and mobile application readouts. c) DIY electrochemical biosensor

ABSTRACT

Feline urine provides valuable information on an animal's well-being, but professional veterinary collection and analysis of urine samples can be intrusive, costly, and infrequent. Electrochemical biosensors recognize biological elements such as pH, glucose and sodium, and have numerous applications, including in medical diagnosis, environmental monitoring, food quality control and drug discovery. This paper presents circCAT: PURRtentio, a litter box system that uses a electrochemical biosensor to monitor analytes in feline urine. We provide the implementation process of the system that consists of a DIY three-electrode biosensor, a potentiostat, a microcontroller, a ToF sensor and a mobile application. A rinsing mechanism is also included to extend the lifespan of the sensors. The system was tested using three separate electrochemistry tests to ensure accuracy, reliability, and applicability. We prepared and compared electrochemical biosensors with different conductive materials for Do-It-Yourself (DIY) electrodes. The second test compared PURRtentio against an industry-grade potentiostat. The third test compared our system against current veterinary standards for chemical analysis using feline's urine samples. Additionally, we conducted a case study with a cat using PURRtentio for 72 hours. Finally, with results from these research and another series of interviews we did with veterinarian experts, we provide implications and future directions of this technology. PURRtentio presents an innovative and non-invasive means to consistently monitor chemistry elements in feline urine, potentially allowing for early detection and management of cat's health conditions.

CCS CONCEPTS

• **Human-centered computing** → **Interactive systems and tools**; **Interaction devices**; • **Applied computing** → **Life and medical sciences**.

KEYWORDS

biosensor, electrochemistry, potentiostat, urinalysis

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1 INTRODUCTION

Feline urine, much like human urine, serves as a valuable diagnostic tool for assessing the health of animals. Urinalysis is a common method employed to detect various small animal illnesses, with a particular focus on lower urinary tract issues in cats [13, 65]. However, obtaining this information can be challenging for both general and specialized practitioners, as it typically requires access to clinical laboratories, involves considerable costs, and entails waiting for results to be communicated back to pet owners [65]. Additionally, repeating tests involves repeating the entire process, which is inconvenient for pet owners. While some veterinarians suggest that full urinalyses are not always necessary, simpler alternatives for specific assessments are currently lacking (as detailed in Section 6). Consequently, this project aims to provide a more time-efficient and readily accessible system for monitoring pet health, complementing the urinalysis conducted in clinical settings.

Although urine dipsticks equipped with colorimetric biosensors, which change color based on analyte concentration, are commercially available and accessible to the public, their accuracy and efficiency remain uncertain. Several analytes measured on common test strips may prove unreliable due to data and environmental factors that can impact results [5, 41, 50]. For example, urine test strip indicators for feline proteinuria have demonstrated poor performance [61]. In comparison, electrochemical biosensors, which we explore in this paper, offer greater reliability than their colorimetric counterparts. Electrochemical biosensors enable real-time monitoring of biomolecules or analytes by measuring changes in concentrations of glucose, sodium, pH, and metal ions. These biosensors rely on electrodes connected to a potentiometric, voltammetric, or amperometric system. They have been designed for various applications, such as smartwatches[9, 89], temporary tattoos [10], orthodontics [78], permanent tattoos [79], and e-textiles [90].

PURRtentio introduces the use of electrochemical biosensors for animal urinalysis. Our design rationale for employing electrochemical biosensors for feline urinalysis is to enable continuous fluid monitoring, provide a rinsing system to extend the biosensor's lifespan, and offer an integrated system with data logging and visualizations. We propose a reusable and continuous biosensing method that employs an electrochemical system to assess feline urine unobtrusively. PURRtentio comprises a DIY three-electrode biosensor, a potentiostat, a microcontroller, a ToF sensor, a mobile application, and a rinsing mechanism. When a cat enters the litter box, the ToF sensor detects its use, guiding the urine along a filtration system path to deposit it on the electrode. The potentiostat reads the potential value on the electrode and transmits it to the web application, which translates it into a sodium concentration value. Concurrently, the system rinses the electrode. In this paper, we focus on evaluating the system's effectiveness for one analyte: sodium levels in feline urine. Sodium levels are ideal for this evaluation because they can significantly vary due to dietary intake, making fluctuations detectable even in healthy cats [17]. Additionally, sodium serves as an indicative marker of feline health [51, 63]. Furthermore, when assessed in combination with other analytes, sodium becomes even more valuable, aligning with our project's goal of multiple analyte analysis in the future. Thus, sodium is a suitable candidate for evaluating our system's performance, both in technical assessments using artificial urine and cat urine samples, as well as in the case study involving actual cats.

This paper presents the following contributions:

- Introducing electrochemical biosensors for animal urinalysis as an unexplored and promising approach for unobtrusively and continuously monitoring feline health. We discuss the implications and future directions of this technology.
- Developing a comparably low-cost, open sourced litter box with electrochemical biosensors and a potentiostat, and proposing a reusable biosensing method that uses DIY electrodes.
- Our evaluation method that not only technically evaluate the DIY electrodes, the system and compared with real feline urine's samples, but followed an in-the-wild approach on a cat's daily environment.

The paper is organized into several sections that detail the development and testing of the circCAT: PURRtentio system. In Section 3, we discuss the system's design and implementation, including the DIY three-electrode biosensor, potentiostat, microcontroller, ToF sensor, and mobile application. We also describe the rinsing mechanism used to extend the sensors' lifespan. Section 4 presents the results of the technical evaluation, encompassing three separate electrochemistry tests: a) the preparation and comparison of electrochemical biosensors with different conductive materials for DIY electrodes, b) a comparison of cirCAT: PURRtentio against an industry-grade potentiostat, and c) an evaluation of the system against current veterinary standards for chemical analysis using feline urine samples. In Section 5, we describe a case study in which cirCAT: PURRtentio was employed to monitor a cat's urine chemistry over a 72-hour period. Finally, Sections 6 and 7 present the implications and future directions of this technology.

2 RELATED WORK

2.1 Urinalysis and Feline Health

In Bovens' analysis of feline lower urinary diseases, various methods for obtaining urine samples were discussed, including cystocentesis, catheterization, free catch, or litter tray collection. Among these, the free catch or litter tray methods are considered the most reliable for assessing hematuria and are less prone to contamination from external bacteria on the cat's body [13]. The litter tray method is particularly non-invasive and more natural for cats, making it preferable compared to other intervention methods. Timely and proper sample collection is crucial for accurate urinalysis results [65].

Several research studies have explored feline urine analysis methods. In Raskin et al.'s work, they evaluated pH measurements at home versus in laboratory settings. They identified differences in effectiveness between digital meters and test strips, as well as variations in the usable time frame of the samples [64].

Typically, urinalysis is not conducted more than once during a veterinary visit. Diagnosis beyond that point relies on symptom monitoring and treatment [17, 50, 65, 91]. The typical method for obtaining a urine sample involves medical intervention rather than regular urination, making our system significantly less obtrusive [54, 65**?**]. While detailed and precise analysis remains necessary, our system could complement veterinary care, promoting owner awareness of cat health and providing valuable chemical insights into the cat's body at home to aid in diagnosis and care.

The concentration of sodium in cat extracellular fluid and plasma is approximately 155 mEq/l [20]. Sodium levels in cat urine can be correlated with kidney and lower urinary health, as mentioned previously [51, 63]. In other species, variations in urinary sodium levels can be related to adrenal gland malfunction, kidney problems, and even heart-related issues [53, 80]. Sodium intake also strongly correlates with urine output, affecting not only its volume but also the sodium concentration, which can vary from 126 to 830 mg per day in cats [63, 87]. Hui et al.'s research found that increased sodium intake had an impact on the concentrations of struvite and calcium oxalate, which, in turn, are correlated with urolithiasis. This suggests that urine sodium concentration, proportional to sodium intake, is relevant to such issues [87]. Overall, selecting feline urine sodium levels as the analyte to study in our project is justified due to their significant variation, ease of manipulation, and relevance to cat health.

2.2 Urinalysis Systems

Research has explored the manipulation of cat litter in various studies. One study aimed to determine if certain chemicals would make litter more appealing to cats [25]. Addie et al. manipulated cat litter with FCoV to identify which types of cat litter would best prevent feline coronavirus infection [2]. Numerous patents have employed colorimetric pH-sensing silica materials in cat litter. These patents use a litmus agent to change the litter's color when the cat urinates on it, allowing owners to determine the urine's pH level [24, 91]. Other patents have explored detection through cat litter using chemical indicators [42]. One notable patent involves using cat litter to detect diabetes in cats, employing an absorbent substrate manipulated with sugar-detecting chemicals [70]. Relevant to our system, a group created a salivary urea sensor using a diode and photo-conductive cell to detect blood urea nitrogen in chronic kidney disease patients [82]. This research was later applied to create a cat litter capable of sensing kidney diseases, similar but different from our biosensor system. However, there is currently no internationally published research on their system.

Maintenance, especially concerning the system's sanitation, plays a crucial role in making it unobtrusive. Research by Severin and Hayes suggests that electrode rinse solutions can improve process efficiency by up to 56 percent. This research specifically used sodium chloride, which is also used in our project [66]. Other previous studies have indicated that electrode rinsing solutions can enhance the longevity and performance of screen-printed electrodes, with the effectiveness varying depending on the materials used [23, 69]. In our project, we implemented a pumping system to rinse the electrode promptly after each use.

2.3 Electrochemical Sensors

Most industry-standard electrochemical electrodes or biosensors are screen-printed pieces made with carbon or silver-silver chloride [37, 39, 45, 67]. However, we aimed to create our biosensors with materials that promote replicability. Studies have explored the use of copper as an alternative [32, 58]. Many studies have employed copper for biosensing purposes and found it advantageous in various ways [32, 36, 38, 48, 58]. The primary concern with using copper is oxidation, but research on the effects of oxidation on copper's conductivity levels indicates that the loss of conductivity is negligible compared to potentiostat noise and sensor lifespan in our scenario [11, 12, 30, 32, 46, 47, 58, 75]. Sensor lifespans for screen-printed electrodes and similar replaceable pieces are estimated to be around 3 days under general circumstances, even with laboratory-grade cleaning and care, typically involving rinsing and storage at below room temperature [4, 18, 33, 35**?**]. Therefore, the depreciation in copper mass to cause significant changes in sensor readings is not significant. To preserve the lifespan of our sensors and ensure reading accuracy, we implemented a rinsing system with a rinsing solution, following laboratory practices.

3 CIRCAT: PURRTENTIO IMPLEMENTATION

3.1 Form Factor: Litter Box

As shown in Figure 1, we adapted a two-layer litter box to contain the required hardware. The bottom layer houses the potentiostat system, a pee pad for fluid absorption, and the rinsing system, which directs rinsing solution from a container accessible from outside the litter box. The bottom layer, specifically the biosensor, receives fluids through a sifting and funneling system for data measurement. Non-absorbent cat litter is placed on a plastic, slanted funneling surface to filter all liquid. A second funneling plate ensures that all liquids are directed to an opening directly above the biosensor. On the upper layer, a distance sensor detects when the cat enters and exits the litter box.

The design aims to be inconspicuous on the outside to avoid drawing unnecessary attention from the cat. It can be powered either by a portable battery or an outlet. Figure 1.b shows the system with the components on the exterior (when using a portable battery, including the pump and cables). None of the electronic components come into direct contact with the cat to ensure its safety and system functionality. The rinsing solution container is located outside the box for easy access but remains hidden from view when the box is appropriately positioned against a wall. We use lightweight, animal-safe products to minimize wire exposure.

3.2 Electrochemical Biosensors Fabrication Process

Electrochemical biosensors are devices that use electrodes to detect and measure biological molecules. In this project, the biosensors are used to detect sodium levels in feline urine. These biosensors work by reacting with sodium in the urine, producing an electrical signal that can be measured by the electrodes. This potential can then be converted into a sodium concentration level and sent to a mobile application for further analysis and monitoring. Electrochemical biosensors are widely used in healthcare, environmental monitoring, and food safety testing due to their high sensitivity and selectivity. They offer a simple, fast, and cost-effective way to detect and measure biological molecules, making them an excellent tool for monitoring analyte levels in feline urine.

An electrochemical biosensor typically consists of three electrodes: a working electrode, a reference electrode, and a counter electrode. The working electrode is coated with a sensing material (sodium activation solution) that reacts with analyte changes. The reference electrode serves as a reference point to measure the voltage generated at the working electrode, with its potential remaining constant throughout the measurement process. The counter electrode provides a source of electrons to balance the current flow in the electrochemical cell.

3.2.1 Biosensor Design Decisions. To design our electrodes and the form factor of the system, we conducted a materials exploration. We considered various materials for the electrodes, including silver (Ag), silver chloride (AgCl), carbon (C), gold (Au), graphite, and copper (Cu) [8, 37–39, 45]. We also experimented with different substrate materials, such as ceramic, paper, adhesive paper, polymer, kapton, and textile.

Ultimately, we restricted our search to commonly used polymer and textile materials suitable for DIY prototyping due to their flexibility, resistance to dissolution, and compatibility with activation by the sodium activation solution. We tested various plastic and fabric materials, including vinyl, ABS, TPU, acrylic, PU leather, nylon fabric, PET, and fleece. Fleece and nylon fabric were ruled out for sanitary and contamination reasons. Sturdier materials did not hold urine samples consistently and performed poorly in testing with undetermined fluid flow rates and amounts. We then tested various conductive materials on thin acrylic sheets, PU leather, and vinyl, including gold leaf, silver paint, carbon paint, graphite powder, and copper sheet tape.

After extensive testing, we found that PU leather on 1.5mm acrylic sheets offered the desired balance of flexibility and structure retention. The dimension of the electrode was determined to be 0.7 inches in diameter in the sensing area, suitable for fluid flow in our system.

The choice of copper tape electrodes with insulating adhesive was made due to their low cost and ease of replication. These electrodes are oxidation-resistant, making them suitable for our application. The oxidation resistance ensures that the rate of oxidation, specifically its effects on conductivity, is negligible. Biosensor lifespan is protected by the rinsing solution, but typical industry electrodes have a similar lifespan primarily limited by chemical reactions or physical material damage from use.

3.2.2 Biosensor Design Decisions. We performed a materials exploration to design the electrodes and the form factor of the system. Before any electrochemical testing, we first needed to craft physically durable and suitable electrodes. We considered many previously applied material choices available, such as silver (Ag), silver chloride (AgCl), carbon (C), gold (Au), graphite, and copper (Cu) [8, 37–39, 45]. For the substrate material, we experimented with ceramic, paper, adhesive paper, polymer, kapton, and textile [59, 60, 83, 85, 86, 90]. We restricted our search to polymer and textile materials that are commonly used for DIY prototyping, and for their flexibility, indissolubility, and capability of being activated by the sodium activation solution. Thus, various plastic and fabrics were tested, including vinyl, ABS, TPU, acrylic, PU leather, nylon fabric, PET, and fleece. An initial test with cat urine in the litter box setting ruled out fleece and nylon fabric for sanitary and contamination reasons. The sturdier materials did not hold the urine samples in place consistently and did not perform well for testing with undetermined fluid flow rate and amount. Finally, we tested our various conductive materials on thin acrylic sheets, PU leather, and vinyl. We used gold leafs, silver paint, carbon paint, graphite powder, and copper sheets tape. PU leather and vinyl retained all materials, while acrylic sheets had some flaking and slipping with certain materials.

After experimenting with acrylic sheets between 0.2 to 1mm, the 0.3mm one did have the most desirable flexibility in terms of maintaining structure while interacting with fluid output from the litter box, but malleable enough to be curved by the heavier pump flow of rinsing solution [88]. Vinyl was somewhat too soft, which resulted in fluids slipping from part or all of the electrode, which would give inaccurate electrochemical measures. PU leather of less than 1mm had this same issue, though lessened. PU leather of 1.5mm performed about as well as acrylic sheets. The test was done with heavy water flow through only the litter box sift and our manipulated funneling system, without additional litter matters, so it is safe to assume that 1) feline urine flow will not be as heavy, and 2) additional sifting will also reduce the force of flow. Our box design and fluid flow through the litter also contributed to determining the size of the electrode (0.7 in diameter in the sensing area), being not too small for the fluid opening, but also not too large as to be prone to an irregular area of contact with fluids.

An additional factor we considered was activation solution and barrier applications. Vinyl and acrylic sheets change more dramatically than PU leather to temperature changes [29, 88]. In addition, PU has shown promise as a substrate in past literature [29]. Due to its cost and manipulation, PU was more convenient for our design considerations.

3.2.3 Electrode Fabrication. Our DIY biosensor fabrication process consists of copper tape electrodes cut by a vinyl cutter and placed on a film substrate, a sodium activation solution applied to one of the electrodes to ensure sodium selectivity, and an insulating barrier solution over the non-sensing portion of the electrodes for insulation. We purchased both solutions from Zimmer and Peacock [**?**]. Our design process is shown in Figure 2.

Electrochemical Biosensor Fabrication. The flexible 3-electrode copper electrode is applied to a clear or mica-colored Polyurethane (PU) substrate. The substrate is an insulating material regardless of color [6, 74], and PU has also been shown as a capable electrode substrate in past research [29]. A home-crafter cutting machine (Cricut) is used to cut the shape and size of the electrode using oxidation-resistant copper tape with insulating adhesive. We attached the electrodes to a PU sheet of 1.5mm thickness, maintaining a hydrophobic semi-flexible body that enough liquid could bend and drip down from, in contrast to other biosensor substrates made from plastic, ceramic, or paper [15, 22, 52]. We also tested with vinyl, fabric, and other materials as substrates, but PU was the one that ultimately was sturdy enough to not fold onto itself and hold its shape, but malleable enough to bend when enough liquid

Figure 2: Electrochemical biosensor fabrication process: a) Copper tape cut by the vinyl cutter. b) Sodium activation solution is applied. c) Barrier solution is applied.

gathered. In addition, the electrochemical properties, in terms of oxidation resistance and chemical reactions, of using copper with PU are desirable [14, 27, 29, 30].

The overall dimension is 0.7 inches wide by 2 inches long, based on the areas of fluid flow in our system, flexibility, and testing. Other aspects of our fabrication process, such as for materials, designs, and making techniques, were also based on many criteria [40, 67, 72]. Our first electrochemical study for this project, detailed in the evaluation section of this paper, describes these considerations comprehensively. Another deciding factor is the amount of pee produced. A cat pees on average 36 mL/kg of body weight per day [7], the average giant cat weighs 4.9 kg, and a cat pees around 2 to 4 times per day [21, 49]. This calculates to 58.8 mL per pee session, about 1/4 cup. Given the rate of flow in our system, we tested with 1/4 cup of fluid to see if the electrode would be exposed amply.

The decision was to use antioxidant-free, oxidation-resistant copper tape [14, 76] made with insulating adhesive. This makes the tape oxidation and corrosion-resistant without affecting electrochemical behaviors [19, 57, 92]. Incidentally, copper tape electrodes are easy to replicate and a cheap material choice compared to typical carbon or Ag/AgCl used for screen-printed electrodes [15, 39, 52]. With oxidation resistance, the rate of oxidation, specifically the effects on conductivity, becomes negligible [58, 71, 81, 92]. The lifespan of the electrode will be protected by the rinsing solution, but the biosensor lifespan will still remain about the same as for most industry electrodes, where the chemical reactions or physical material damage from use are the main impediments [4, 35, 44].

Sodium Activation Solution. We used a sodium activation solution (from Zimmer and Peacock) [?] on the working electrode to turn the electrode into a sodium biosensor. The activation solution reacts with the working electrode to make the biosensor sodium ion-selective. The solution also ensures that other elements exposed to the biosensor do not affect the results.

Barrier Solution. We then covered the rest of the exposed electrode areas with insulating barrier solution [?] to ensure that the conductive leads prone to solution exposure are insulated. This is so that only the biosensor portion will be in contact with solutions for precision in measuring.

3.3 Hardware and Software

3.3.1 Electronics: The base hardware consists of an Adafruit Feather M4 Express board ($25), an Adafruit Bluefruit LE SPI Friend module ($18), a VL53L0X Distance sensor (ToF) ($15), and a 3V submersed fluid pump ($3). The potentiostat we are using for our project is a Rodeostat Featherwing V0.3 R1 potentiostat ($23) from IO Rodeo [1]. This is an inexpensive, conveniently sized potentiostat with 1000uA capacity. In order to make it compatible with our goals and other hardware, we modified the circuitry and created our own software implementation.

To initiate the testing process, the Time of Flight (ToF) sensor detects when a cat enters the litter box and begins an Open Circuit Potential (OCP) test once the cat has finished using the litter box and fully exited. OCP is the voltage present when the terminal ends of a circuit are detached and can be used to measure analytes such as sodium. This allows the urine to pass through the litter, sift, and funnel, providing ample time for the biosensor to stabilize before running a test. Research indicates that cats typically spend approximately 1 minute loitering in the litter box before urination, up to 1 minute urinating (in ill cats or extreme scenarios), and up to 1.5 minutes loitering post-urination in non-clinical settings [21, 49, 56]. On average, about 1/4 cup of fluid passes through to the biosensor, enough for a test in approximately 38 seconds, as tested with water. From the chosen testing time period, it is evident that our biosensors are capable of producing stable data for at least up to 100 seconds post-exposure, as shown in Figure 8. Our system ensures that each OCP test occurs when the sensor is stabilized and the system is ready[2].

Our testing process operates asynchronously with data communication. We configured the system such that the sampling rate is well-suited for biosensor behaviors, as detailed in our evaluations,

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[1] https://iorodeo.com/

[2] https://github.com/anonpapersandsuch/purrtentio

and to account for the time needed for Bluetooth Low Energy (BLE) communication with our mobile application.

3.3.2 Rinsing System. We implemented a rinsing system to enable continuous and repeated measurements. Drawing from previous literature, various methods exist for revitalizing biosensors to maintain their shelf life and precision [23, 62, 66, 69]. For our project, we procured a suitable biosensor rinsing solution from ZP[3]. Our submersed pump exerts sufficient force to rinse the biosensor after testing. The pump operates for 3 seconds, dispelling enough fluid to flush the urine and partially immerse the biosensor for chemical cleansing before drying, which takes approximately 10 minutes. It is worth noting that a cat is unlikely to urinate again within that time frame [7, 21, 49]. Furthermore, a quarter cup of fluid passes through the system in less than 130 seconds, so rinsing does not commence until the urine has completely passed through.

Testing using our system remains inaudible and unobtrusive. However, when the rinsing system is activated, it does produce some noise, measuring at 60 dB, which is considered low for both humans and cats [31]. Additionally, the pumping occurs a while after the cat exits the litter box, following the aforementioned time frame. Therefore, this should not significantly impact the discreet and unobtrusive nature of our system.

3.3.3 Mobile Application. To facilitate the visualization and storage of urine data, we developed a web application that establishes a connection to the device via a BLE module and utilizes the p5ble.js library. This web application offers real-time data visualization through a live graph and employs a gauge to visually represent the concentration value range. The use of color in the visualizations serves as an indicator of whether the concentration falls within the normal or abnormal range. Testing data from the sample is transmitted as the electrochemical test occurs. After each measurement, we send the data to the application for calculations, and the hardware resumes the next measurement until testing is complete. Once the test finishes, the application updates the most recent urine analysis log for litter box usage.

Our application is designed to provide a user-friendly depiction of the analyte concentration level for the cat owner. The visualization includes the most recent litter box usage event, the estimated concentration, and other electrochemical data (such as potential and current). This feature opens up numerous possibilities for monitoring litter box usage status, extending beyond electrochemistry[4].

4 TECHNICAL EVALUATION

4.1 Validation of Biosensor

We fabricated 5 biosensors with various conductive materials listed in Table 1. The materials we used were based on literature suggestions and availability. Au refers to gold leaf with proper metal leaf adhesive. AgCl refers to paintable silver/silver chloride ink. C refers to paintable carbon ink. Cu refers to copper tape. We also attempted with printable silver nanoparticle ink as most SPE and past research uses [37, 39, 45, 67], but it did not work properly with our adhesives, and the paintable inks did not retain well enough for most of the substrates we attempted, namely sticker paper, PU, acetate.

Figure 3: PURRtentio App UI

Table 1: Electrodes Material

	Working and Counter	Reference
AuAgCl	Au	AgCl
AgCl	AgCl	AgCl
CAgCl	C	AgCl
CuAgCl	Cu	AgCl
Cu	Cu	Cu

To test the 5 DIY biosensors, we exposed each of them to the same set of sodium calibration solutions. We also included a control industry SPE, obtained from ZP. In order to ensure validity of the potentiostat, for this step, we tested with EmStat using Open Circuit Potentiometry (OCP) for all trials.

We used previous research for referencing the range of values that sodium levels can vary in feline urine [17, 51, 87]. As with any electrolyte, the range of variance is large, going from 0 to over 300 mM [17, 53, 77]. Given the significant variability in electrolyte levels, we established a safe estimate of the normal range as 20 mM to 260 mM. Based on these criteria, we designed our electrode range capability and precision testing, incorporating ample buffer for both low and high extremes. The concentrations we selected for testing were 0, 34.22, 102.67, 127.48, 154, 205.34, 290.9, and 342.2 mM. To ensure the integrity of our solution preparation, we used preferred techniques and unit conversion [27, 68], which meant favoring validity over evenly distributed, visibly neat values of concentration samples. We prepared the testing solutions through dilution of a highly concentrated solution with known tested value in deionized water. Solutions were mixed by machine controlling speed, time, temperature, and amount [27, 34, 68, 72]. Additionally, we used colorimetric test strips to roughly confirm the validity of the concentration in the prepared solutions [3, 5, 55].

Overall, we found that while the commercial-grade sodium-activated sensor (ZP) worked most reliably, our copper ones also performed comparably. We noticed oscillation of data from all the electrodes using the system. This behavior is an expected outcome

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[3] https://www.zimmerpeacocktech.com

[4] https://github.com/anonpapersandsuch/purrtentiowebapp

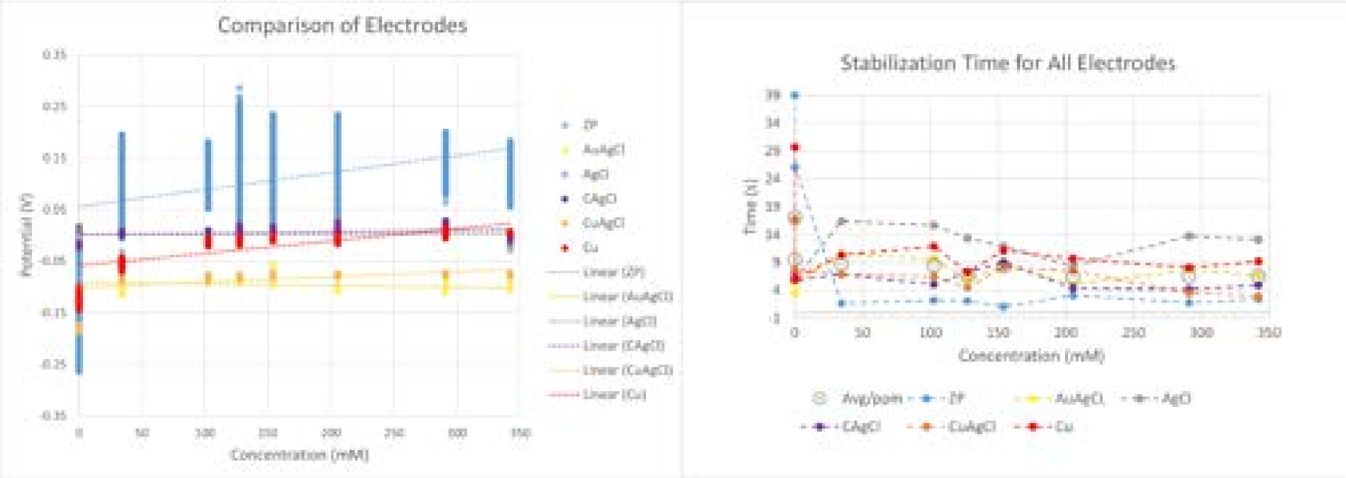

Figure 4: a) Comparison of the potential for each electrode design in varied concentrations of Na. b) Stabilization time of each electrode design in varied concentrations of Na.

due to limitations in the potentiostat hardware, and the sampling rate [16, 83, 84]. To gain a better understanding of the data quality, we examined both the oscillation amplitude and the standard deviation, considering them as indicators of "noise" generated by the electrodes. The final criteria we looked at are the following:

- Trendline reliability for predicting concentration
- Stablization time from when the electrode is exposed to fluid to when it is able to read a stable potential
- Oscillation amplitude
- Noise/ Standard deviation
- Lifespan

The trendlines and stabilization time for each electrode based on each solution concentration are shown in Figure 4. We can see that the commerical electrode (ZP) had the best trendline for predicition, but CuAgCl and Cu performed similarly in slope, only with a different offset. The r squared value for Cu was the highest of all electrodes, even control. We also see that ZP had the most oscillation and noise for the setting of this project. These data are raw inputs and outputs without software stabilization, calculations, or further noise filtering techniques. Due to the predictable sinus behavior of oscillation, we then altered our design to cater to that even if we are limited by hardware to reduce the behavior altogether. To do this, we looked at this criteria on top of noise. Data on the results are in Table 2.

After considering the electrochemical results and taking ease of design into account, we concluded that copper is the most suitable solution for our project. Thus, we made improvements to our hardware and software design to filter our data to the ideal prediction model. Like when testing with the EmStat, our system using Rodeostat also produced the expected oscillation and noise. We altered circuitry of the potentiostat to smooth the data measuring process as much as possible for our specific system, knowing the power supply and components used. We took into account the nature of OCP to remove unnecessary workload on counter electrode or for voltammetric features that does not apply [15, 83, 84]. We were able to reduce noise and mitigate oscillation. We further treated the oscillation by manipulating data calculation and sampling rate to consider the wavelength of the curve, as to use the average point as a point of reference, while still taking into account where the actual data lies.

4.1.1 *Reusability.*

We assessed each electrode’s lifespan by determining the number of reliable tests it could perform. Standard electrode and chemically activated biosensors typically last about 3.5 days, influenced by factors like cleaning and storage conditions [1, 4, 26, 73]. Therefore, our goal is to determine if the electrode can last for the full 3.5 days without experiencing external physical damage or other factors that could shorten its lifespan. Research states that cats urinate 2 to 4 times per day on average, with extremes, in cases of ill cats, of up to 6 times per day [21, 49, 56]. To simulate our expected use case, we conducted tests on each electrode for up to 30 uses, even though our projected usage rarely exceeds 21 uses. Unfortunately, the Au electrode didn’t meet durability criteria due to gold leaf material flaking and peeling. Additional adhesive would have compromised its conductivity, making gold leaf unsuitable. Additionally, the AgCl electrode began producing varying offset values after approximately 25 uses, rendering it unusable.

4.2 Validation of Electrochemical System

In a second study, we compared our modified system, using the affordable Rodeostat from IORodeo, with EmStat, a more costly industry-grade potentiostat. EmStat employs higher-cost hardware with enhanced precision components compared to the Rodeostat. Figure 5 depicts a concentration level tested using cirCAT: PURRtentio, with the ZP electrode as a control and our Cu electrode introduced. We tested solutions with concentrations of 43.48, 97.83, 156.52, 195.65, 234.8, and 391.3 mM, prepared and used following previous procedures. This time, we focused on testing system capability and precision within a closer range of concentration levels. Results are shown in Figures 6 and 7.

Table 2: Electrodes Testing Results

Electrode	slope (v/mM)	r squared	oscillation (v)	noise (v)	lifespan (uses)
ZP	0.00031	0.68	0.01	05	30+
AuAgCl	-0.00006	0.62	0.0009	0.0002	21
AgCl	0.00001	0.39	0.000168	0.002	30+
CAgCl	0.00009	0.62	0.0008	0.003	25-26
CuAgCl	0.00018	0.79	0.0006	0.0018	30+
Cu	0.00027	0.72	0.002	0.003	30+

Figure 5: Testing cirCAT: PURRtentio with different concentration levels

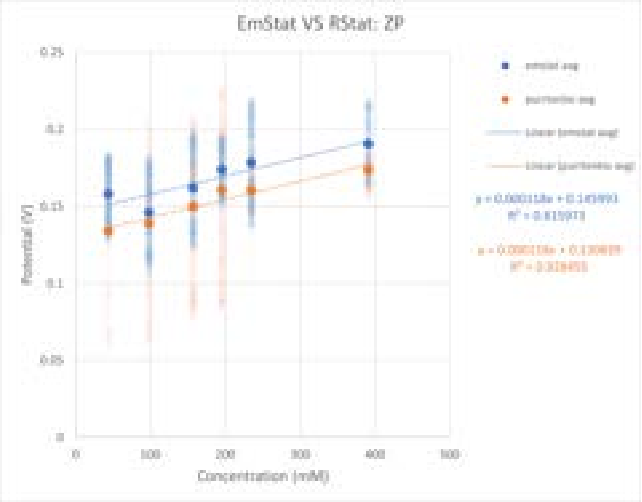

Figure 6: Comparision of EmStat and purrentio using RodeoStat, using ZP sodium sensor

During this phase, we exercised control over the sampling rate, initial current, and offsets induced by the hardware (excluding the biosensor). Consequently, we observed improved trendlines with EmStat compared to previous tests. Notably, our system exhibited very similar results regardless of the electrode used. While our system had lower oscillation than EmStat, we attribute this difference to EmStat's higher sensitivity due to the use of more expensive production materials and methods. EmStat is designed to excel in a variety of electrochemical projects [14, 34, 39, 83]. Overall, our system compares favorably with the commercial product based on our testing objectives. This is evident from trendline analysis and the r-squared value (as shown in Figures 6 and 7). The slope values are remarkably similar down to the ten-thousandth place, and the r-squared values are within 0.1 of each other, demonstrating the strength of the correlation between measured values and expected values.

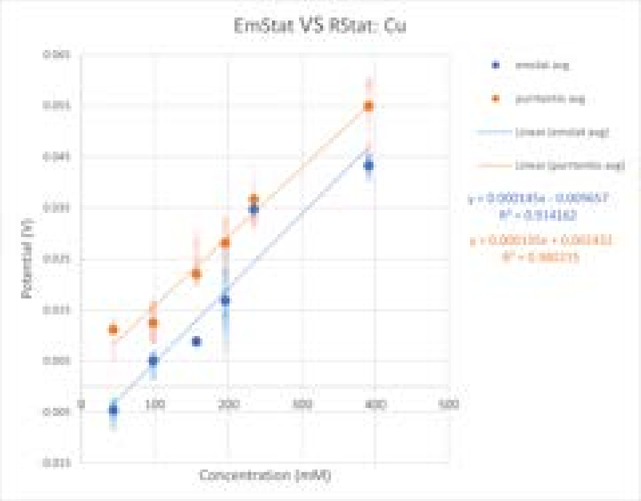

Figure 7: Comparision of EmStat and purrentio using RodeoStat, using copper sodium sensor

4.3 Evaluation with Feline Urine Samples

To validate our system and biosensor, we tested real urine samples and compared our results with current feline urine sodium testing standards. In this third study, we collaborated with a veterinary researcher who provided us with 5 urine samples collected for various purposes. No additional samples were collected solely for our testing, ensuring ethical resource use. Prior to receiving the samples, each underwent laboratory-grade sodium testing at the veterinary facility.

We conducted tests using our PURRtentio device, performing 10 data measurements for each sample and repeating the process three

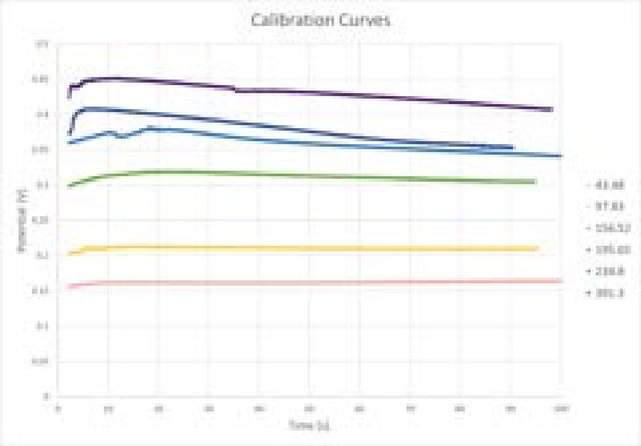

Figure 8: Stabilization curve at various concentrations

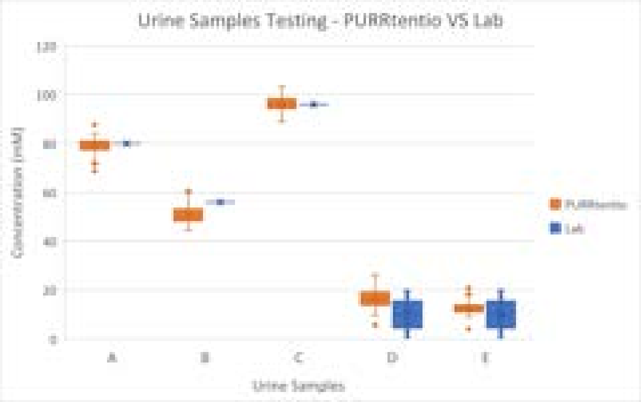

Figure 9: Testing urine samples of various sodium concentration with PURRtentio and standard lab testing

times. Due to limited sample volume (less than 5mL per sample), we employed this approach.

Our results closely matched the laboratory findings. In 75 percent of cases, our precision remained within 3mM of the lab standards, with a maximum deviation of 5 mM from the lab results. Additionally, we successfully measured data below 20mM, whereas the lab results only categorized samples 4 and 5 as less than 20, without specifying exact values. These findings are depicted in Figure 9.

This underscores the accuracy and reliability of our PURRtentio system in measuring sodium levels in feline urine, demonstrating its potential as an effective tool for real-world feline health monitoring.

5 CASE STUDY

We conducted a 72-hour case study involving a 10-year-old cat participant using cirCAT: PURRtentio. Our study qualified for an exemption from IACUC review. The participant is in good health and mildly overweight. To familiarize the cat with the litter box, we utilized the system for three days before the testing period. All usage of the system was voluntary, and alternative litter boxes were available to the participant.

Table 3: Cat Urine Data

Day	Time	Level (mM)
1	6:04AM	29
1	1:00PM	132
2	11:00AM	87
2	5:40PM	40
3	12:59PM	19

Figure 10: User Study: Feline Urine analyzed with cirCAT: PURRtentio

Upon the cat's adaptation to the litter box, we introduced cirCAT: PURRtentio by incorporating electronic components into it. The cat did not display any signs of additional interest or irritation. During the 72-hour testing period, the participant used cirCAT: PURRtentio a total of six times. We measured her sodium levels, as shown in Table 3. As expected, sodium levels varied based on feeding times [53], with elevations observed after the meal at 12:00 PM. Although the next feeding time was at 6:00 PM on the first day, the participant did not choose to use the system after that time. Utilizing existing security cameras, we verified that the usage log in our system corresponded to the cat's activity in the litter box. cirCAT: PURRtentio performed as expected, with no observable abnormalities from our participant. This case study underscores the usability of our system and mobile application in real-world settings.

6 EXPERTS INTERVIEW

6.1 Protocol

An additional research we contribute is a series of interviews. We inquired with veterinary experts in the field on the plausibility and potential of this project. This study provided us with professional affirmation, knowledge, and future direction. This minimal risk expert interview study was determined to not require IRB review.

We formulated six questions to use in semi-structured interviews conducted on seven recruited participants. The participants were English speakers over the age of 18. They have had at least six months of clinical experience with cats in addition to having veterinary credentials. The interviews were conducted online. Audio recordings and transcriptions were saved for later analysis then deleted upon completion. The six questions were aimed at determining the following:

- Impediments to the viability, reliability, or validity of a continuous urine monitoring cat litter system
- Use cases for a continuous urine monitoring cat litter system
- Analytes or combinations of analytes that would be useful or insightful for well-being
- Volatility of sodium levels in feline urine,such that sodium would be a useful analyte for demonstrating concentration fluctuation
- Usefulness of sodium level determination in feline urinalysis, such that knowing the concentration would be insightful for well-being

6.2 Findings and Discussion

We were able to gather that electrolyte levels vary greatly, which is convenient for testing our biosensor and system development. Sodium is one of the electrolytes analyzed during typical urinalysis. The interviews matched past paper conclusions [53, 77]. Thereby validating our choice of analyte for this stage of the project: proof of concept, technical evaluation of the system and notice fluctuations in analyte using feline urine samples and an actual cat participant. 2 of our participants mentioned how the project could benefit diabetic cats if suitable analyte or multiple analytes were assessed.

We also learned that the typical method of obtaining a urine sample involves medical intervention rather than regular urination, highlighting the non-invasiveness of our system. As stated by 3 of our participants, while a detailed, precise, holistic analysis is still necessary, our system could serve to complement veterinary care. A participant said that "this (system) would be good to use as an at home follow up." Owner awareness of cat health would be encourage. The chemical status of the cat's body at home would also be a helpful addition to aid in veterinary diagnosis and care. One of the interviewed participants suggested that continuously monitorization of urine could pose possibilities for studying filtration of drugs or urinary excretion of drugs/compounds.

This research provides crucial insights for our project's future directions. While urinalysis is typically performed once during vet visits, our system allows continuous monitoring, which can be particularly valuable in cases requiring multiple tests, such as UTIs, and for assessing conditions like diabetic ketoacidosis and proteinuria. It also offers a way to measure urine concentration without a full urinalysis.

7 LIMITATIONS AND FUTURE WORKS

While our study has shown promise and the potential for cirCAT: PURRtentio, several limitations require addressing, opening doors for future enhancements. Currently, our sodium biosensor, while effective, has a limited scope, primarily focusing on specific illnesses. To broaden its diagnostic utility, we plan to incorporate additional analytes, such as glucose, commonly monitored in diabetic cats. Integrating multiple biosensors will provide a more comprehensive view of feline health, aligning with specific illnesses.

In veterinary settings, sterile conditions are essential for urine sample collection to ensure accuracy and prevent contamination. To address this, we'll implement a filtration system within our sifting mechanism.

While our study provided valuable insights, it involved a single cat participant. Future research should include a larger participant pool, including scenarios in rescue facilities. To support this, we'll integrate an RFID reader to monitor multiple cats' health.

In upcoming projects, we aim to integrate cirCAT: PURRtentio with common household IoT devices, such as smart feeders, water intake controllers, and pet video trackers. This integration will enable real-time data analysis for feline diabetes management. Pet owners will receive tailored alerts and medication recommendations based on biosensor data, simplifying diabetes care and improving feline quality of life [43].

8 CONCLUSION

We introduced PURRtentio, a novel approach for unobtrusive and continuous monitoring of feline health using electrochemical biosensors for urinalysis in a litter box system. We developed a cost-effective solution by utilizing DIY electrodes and a low-cost potentiostat, thus promoting replicability of hardware and software. To assess the efficacy of our approach, we conducted technical evaluations of the electrochemical electrodes that tested the performance of the DIY electrodes, system, and comparing them with real feline urine samples. Moreover, an in-the-wild study was conducted in a cat's natural environment over a period of 72 hours. Veterinary experts were interviewed to assess the potential and implications of this research in their practice.

Electrochemical biosensors have demonstrated their effectiveness in human body fluid monitoring and have been integrated into various wearable devices, such as smartwatches, temporary tattoos, and e-textiles. Given the similarity of analytes found in feline and human urine, this project has the potential to inform future research in human health's monitoring. Moreover, PURRtentio represents a valuable solution for pet owners and veterinarians, enabling them to continuously monitor and manage feline health conditions in a non-invasive and uninterrupted manner.

We envision several promising directions for this technology. Future works will expand the range of analytes that the system can detect, incorporating multiple biosensors capable of simultaneously measuring different analytes. Moreover, larger-scale testing will be conducted to further validate the system's performance and reliability. Integration with other IoT devices, including smart feeders and activity trackers, will enhance its functionality and provide a more comprehensive view of feline health. Additionally, by leveraging machine learning techniques, we can enhance data analysis capabilities and enable the prediction of health conditions based on collected data. Furthermore, given the similarity of analytes found in feline and human urine, our project holds potential for influencing future research in the field of human health monitoring.

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