Team Results Document
UCTeam

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SensUs

SensUs 2024

Acute Kidney Injury

Table of Contents

1. ABSTRACT

Acute kidney injury (AKI) is a condition characterized by a rapid decline in renal function. Assessing levels of creatinine, a key biomarker for kidney function, can facilitate the early detection of AKI and expedite treatment.

UCTeam from University of Chemistry and Technology in Prague presents a continuous biosensor designed to detect creatinine levels, utilizing a three-enzyme system comprising creatininase, creatinase, and sarcosine oxidase. The biosensor, consisting of two flow cells, contains immobilized enzymes to prolong the stability of the system. Electrochemical detection using screen-printed electrodes (SPE) aims to provide real-time monitoring of creatinine from interstitial fluid.

By monitoring creatinine levels, this biosensor has the potential to enable earlier diagnosis of AKI compared to existing methods. Early detection of AKI is critical, as it allows clinicians to intervene sooner and potentially prevent further kidney damage.

2. Biosensor

2.1 Molecular recognition

In our biosensor, we have decided to use a three-enzyme system – creatininase, creatinase and sarcosine oxidase. During the reaction cascade, creatinine is converted, with one of the final products being hydrogen peroxide. Enzymes were immobilized using glutaraldehyde in combination with BSA to form so-called "Cross-Linked Enzyme Aggregates" (CLEAs) [1]. Addition of BSA aids in the preparation of CLEAs, particularly when the enzyme concentration is low or when glutaraldehyde, necessary for aggregation, negatively affects the enzymatic activity [2]. By immobilizing enzymes, their stability is improved, and the shelf life is extended.

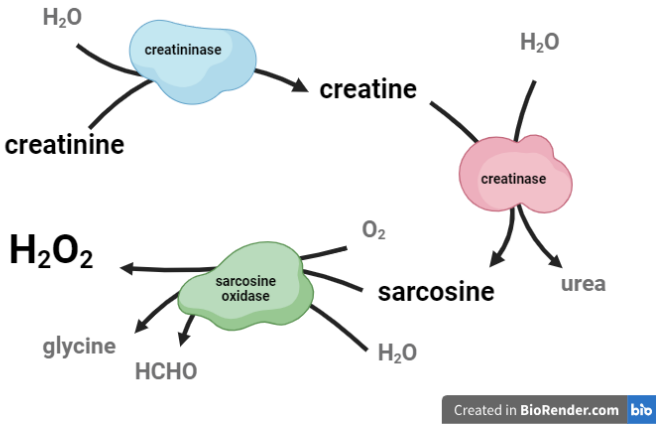
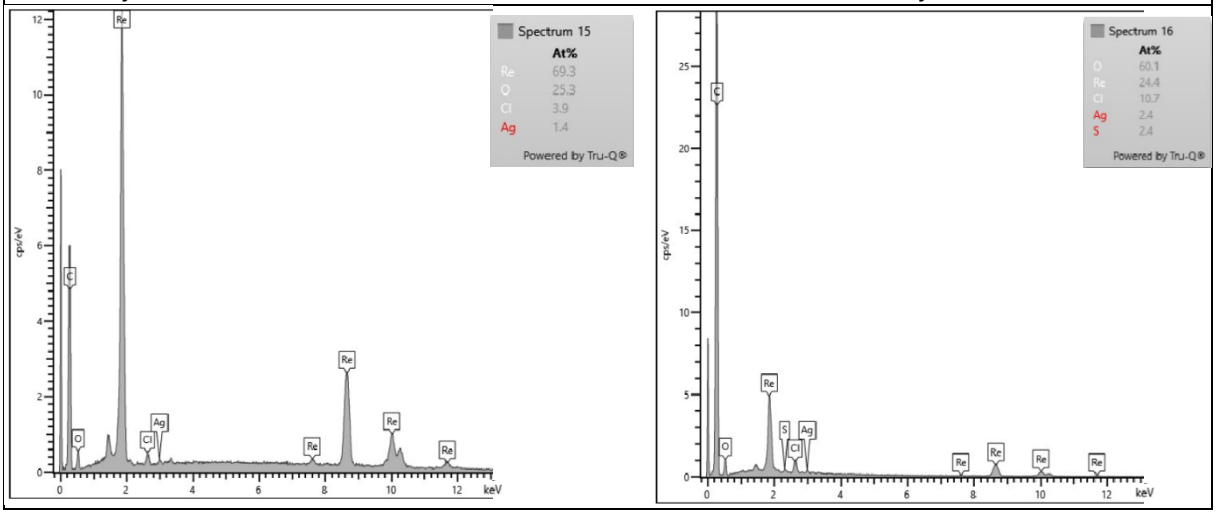


***Figure 1**: Visualization of the molecular recognition.*

2.2 Physical transduction

As the final product of our enzymatic cascade is hydrogen peroxide, electrochemical detection was chosen, as it is well defined in literature. For the simplicity of instrumentation and its sensitivity, chronoamperometry (CA) is our first-choice detection method. Using constant reducing potential during the CA then allows the change of the mediator, ReO_2, from Re(IV) to Re(III), which is then reoxidized by the reduction of H_2O_2, creating H_2O and consuming an electron that is then detected by the electrode [3]. The concentration of creatinine is then calculated by the stoichiometry of the enzymatic reaction and the amount of peroxide detected on the electrode.

EDS analysis of ReO_2 modified electrode surface – C omitted as it is the base layer material



2.3 Cartridge technology

Our biosensor comprises a casing, potentiostat and two cartridges, which are then divided into a flow channel, membrane and a compartment. The body of the cartridges is made of two 40 mm thick PMMA sheets, sealed by a 0.25 mm thick Liqcreate Flexible-X gasket. One sheet houses the flow channel, the other is used mainly as an anchor for the screw connection. The measuring compartment is then created by a commercially available cellulose dialysis membrane, which separates the main flow from the compartment, and by the gasket, which creates its volume. KidneyGuard uses dialysis membranes to assure impermeability to larger molecules, to keep enzymes from washing away, and also by limiting flow near the electrode, it increases its longevity. At the bottom of the measuring chamber, between the anchoring PMMA and the gasket, the screen-printed carbon electrode (SPCE) is inserted, and the chamber is filled with supporting electrolyte. The second cartridge houses a bioreactor. Its chamber is filled with immobilized enzymes and supporting electrolyte; otherwise, its design is very similar to the other cartridge.

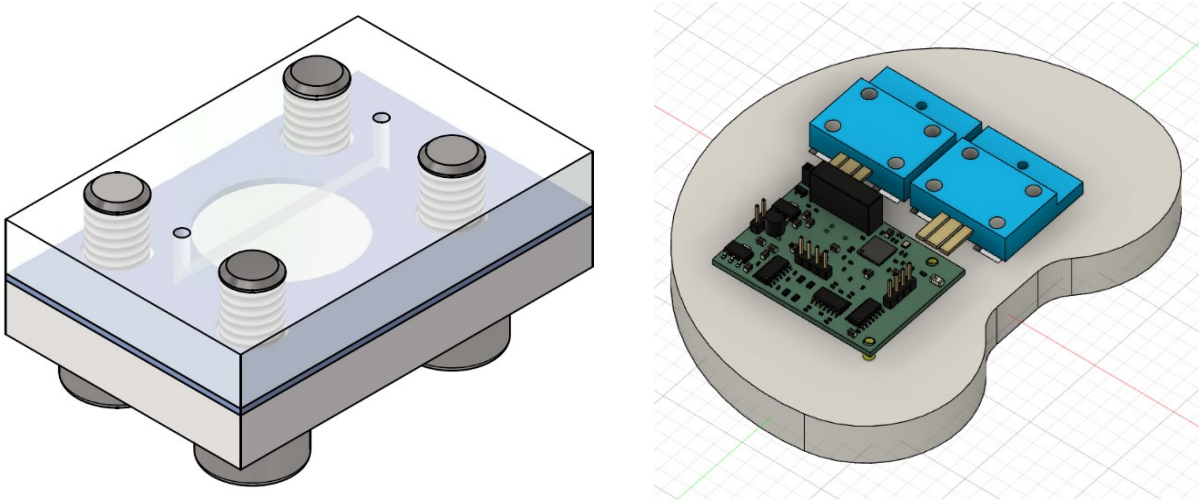


***Figure 2**: Visualization of the cartridge and the bottom part of the casing with potentiostat and both cartridges.*

The SPCE is printed on 0.20 mm polyester support, the contact lines and the reference electrode (REF) are made of silver ink (Dupont 5064), the electrode area was then created by printing over the silver ink by graphene nanoplates (GNP) ink, creating the working (WE) and counter (CE) electrodes. The WE is then modified by drop casting of ReO_2 solution on its surface [4].

2.4 Reader instrument

The sensor instrument measures 50 x 36.5 mm and is powered by a battery. It supports two channels for separate measurements, using a default two-electrode system. Additionally, a plugin was developed for incorporating a reference electrode if needed. The design of our potentiostat was mainly influenced by the works of Meloni [5] and Gao et al [6]. The sensor operates with voltage settings ranging from -1 V to 1 V in 0.5 mV increments and current measurements from -460 µA to 460 µA, with an approximate resolution of 14 nA, not considering noise. These precise measurements are achieved through a four-layer PCB, high-resolution ADCs and DACs, and several operational amplifiers in the analog circuitry. Additionally, there is an option to measure on two channels simultaneously.

At the core of the device is an STM32WB microprocessor with a wireless interface. Communication with a computer is facilitated via a UART interface, allowing user to choose between chronoamperometry or voltammetry, set measurement ranges, and receive data. While a Bluetooth antenna is included in the PCB design, it is currently non-functional due to an issue with trace impedance matching. The goal is for the Bluetooth antenna to connect with a mobile application for user interaction (see **Appendix 12**). The device also features a low-pass FIR filter in the software to eliminate high-frequency noise.

3. Technological feasibility

Our sensor utilizes many attributes of previously created biosensors, as a major inspiration during its development was the glucose sensor, which operates on similar principles. With the cooperation of the University of Pardubice (UPCE), we were able to create a sensitive H_2O_2 electrode with a reproducible response of 60 nA per 30 µM of H_2O_2. Initially, we planned to use two-electrode setup, but during the development, we decided to change electrode material and without reference electrode the signal became close to illegible. The choice to use CA was made mainly for 3 reasons – response sensitivity, low energy consumption and simple instrumentation. Our cartridge features a straightforward design but is made to be easily modifiable. Although its design is not revolutionary, we at least decided to focus on parameters such as – biocompatibility, sustainability, workability, stability and inertness.

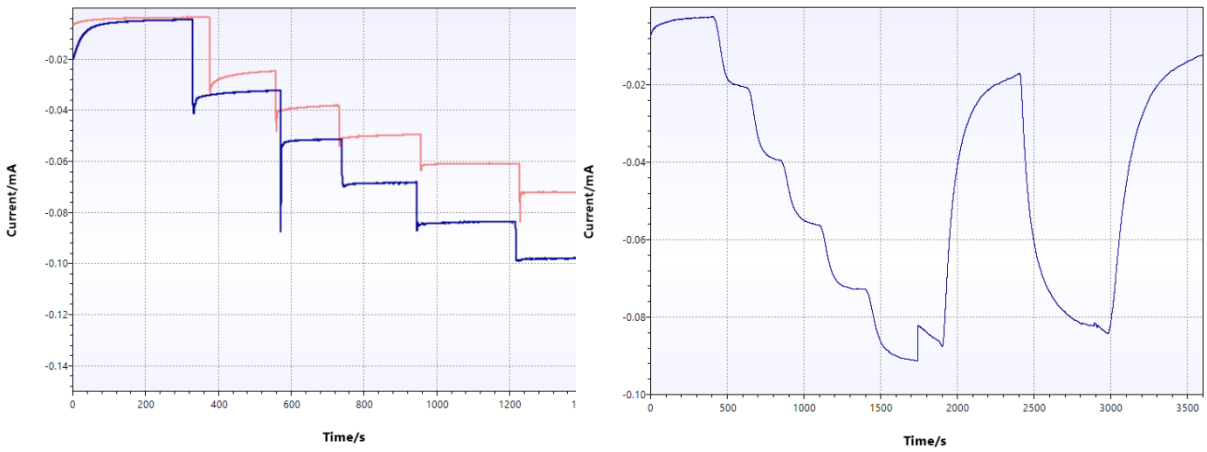


***Figure 3**: Calibration curves (2 repetitions) for millimolar concentrations of H_2O_2 on ReO_2 modified SPCE and measurement showing the regeneration of the electrode in flow system (increase at 1700s is due to a manipulation mistake).*

Our biggest risk is the decision to develop a potentiostat tailored to our system. Currently, it is able to run two different measurements at the same time, either in two- or three-electrode setup. Both its current and potential ranges are serviceable for use in this biosensor, as is its resolution. The main weakness of our system lies in its molecular recognition, as more time would be needed to fully test the shelf life of immobilized enzymes and preservation of their activity. One of our current challenges is ensuring the flow of analyte, which is currently solved by an external intake (syringe pump). This could be resolved by using a smaller pump that could be housed inside the biosensor casing, along with a detachable waste compartment. Another limitation is the current version of the potentiostat. At this point, we are using a 16-bit analog-digital converter, which, at current ± 460 µA range, limits our smallest step to 14 nA without noise. A straightforward solution is using higher-resolution ADC, granting us at least ten-fold increase in resolution. In general, the remaining problems could be solved with more time for optimization. Yet, considering all aspects of the biosensor developed by our team, we believe it has potential to achieve analytical performance in ISF.

3.1 Future plans

Our initial product will be a biosensor utilizing electrochemical recognition based on an enzymatic cascade reaction. During this period, we plan to develop a multiplex point-of-care device to monitor multiple biomarkers, including Cystatin C, NGAL, and Creatinine, for better, earlier, and more accurate recognition of kidney issues. Looking ahead, our vision includes creating a biosensor capable of multi-disease monitoring, incorporating various biomarkers and enzymes to detect multiple conditions simultaneously, enhancing comprehensive health monitoring within a single device.

During the development of KidneyGuard, we tested a few electrode materials, and for the final design, we settled for graphene nanomaterials. In the future, we are planning to test different inks to find material with similar surface enhancement yet better reproducibility. The same optimization is planned for the cartridges. During the development, PMMA proved to be a great choice for its transparency, malleability, recyclability

and inertness when in contact with bodily fluids. Yet, in the future, we plan to focus our efforts on finding material that would have better balance between stability and degradability.

Regarding the enzymes, our aim is to further study their long-term stability. We will focus on finding the optimal storage conditions and advance in finding optimal immobilization technique for our purposes. Besides that, we want to explore the behaviour of the enzyme system under more realistic conditions, for example, higher temperatures or the presence of interstitial fluid over a longer period.

In the current device, the dimensions of the casing and cartridges are enormous, mostly for better manipulation during the placement of components. However, for the future generations of the KidneyGuard, miniaturization is in order, as we hope to build a wearable device. Another possible solution to our external intake problem, which would go hand in hand with our miniaturization effort, would be the use of capillary forces to ensure the flow instead of a pump.

The current potentiostat uses a DC-DC switch mode power supply to achieve symmetrical power, but this introduces a high-frequency ripple. Additionally, several linear voltage regulators are used for multiple voltage levels, which are not very efficient. In the next generation, the power supply design will be simplified to increase power efficiency and reduce PCB size. The issue with the Bluetooth antenna will also be corrected. Furthermore, the next version will include a hardware low-pass filter on the transmitting side to reduce noise on the working electrode.

4. Originality

4.1 Team captains: Ing. Daniel Křížek & Bc. Eva Vogelová

Due to a lack of biosensors-focused groups at UCT and our team members being first-timers in this competition, we did not have any preexisting knowledge and products to build on. These circumstances hindered our development, but at the same time served as a breeding ground for novel and untested ideas. After testing multiple concepts for molecular recognition and physical transduction, our team decided to pursue an enzymatic cascade as a recognition element and chronoamperometry for detection in our final device.

The enzymatic cascade consists of creatininase, creatinase, and sarcosine oxidase and these enzymes are immobilized in the form of Cross-Linked Enzyme Aggregates using glutaraldehyde and BSA. The final product of this cascade is hydrogen peroxide which then reacts with redox mediator ReO_2 deposited on the surface of the working electrode. The electrodes used in our sensor are graphene nanostructured SPEs and were not developed by us, but were acquired from a research group at UPCE. The electrochemical measurements are made possible by a potentiostat fully developed by our team member that enables the sensors to take two-channel measurements with both two and three-electrode setups.

Our biosensor takes the physical form of a casing, two cartridges, and the potentiostat. The cartridges are linked to each other, and the outlet of the first cartridge serves as the inlet of the other one. Each cartridge contains a flow channel and a cell separated by a membrane. The first cartridge's cell contains the Cross-Linked Enzyme Aggregates serving as a bioreactor and the cell in the second cartridge contains the SPE with the redox mediator. This arrangement is very versatile and enables the option for multiplexing.

We believe this combination of new and already established techniques in biosensing produces a highly adaptable and original biosensor.

4.2 Supervisor: doc. Ing. Zdeněk Slouka, Ph.D.

On announcing the topic of the competition, which was continuous monitoring of creatinine, the research team immediately realized they could not utilize any biosensing technologies available at UCT Prague as a starting point. Upon discussing the possible approaches to tackle the problem, the research team performed a thorough literature review focused on possibilities (i) of how to ensure the detection specificity to creatinine and (ii) how to transduce the specificity into a readable output. The team briefly tested various options, eventually choosing to follow a functionally modular concept realized on a microfluidic chip, providing (i) sample pretreatment by a size-selective membrane, (ii) specific recognition of creatinine through a cascade of enzymatic reactions, and (iii) detection through an electrochemical readout. The team embarked on testing and developing each functional module independently when using glucose as a cheap model analyte to detect via enzymatic reaction. Going through many cycles of system development and integration, the team successfully constructed a functional prototype, continuously monitoring glucose concentrations, and confirmed the viability of the chosen method. Unfortunately, the experiments with the enzymes for creatinine detection were delayed due to unexpected issues with their commercial availability despite promises from large companies such as Merck to provide the ordered items by late spring 2024.

Although the main directions of the biosensor development were set in discussions with the supervisors and by confronting various literature sources, the team showed unbelievable determination and ingenuity in realizing the individual steps needed for constructing the functional biosensor. They built their own potentiostat, designed and built the microfluidic platform, and optimized its geometry to accelerate the target detection and quantification. The product of their effort is a continuous, versatile biosensor that will be further tested in the supervisor's laboratory.

doc. Ing. Zdeněk Slouka, Ph.D.	**Ing. Daniel Křížek**	**Bc. Eva Vogelová**
Team supervisor	*Founder; Team captain*	*Team captain*

5. Translational Potential

5.1 Acute Kidney Injury (AKI) Overview

Acute kidney injury (AKI), also known as acute renal failure (ARF), is characterized by a rapid decline (within a few hours or days) in the kidneys' ability to excrete waste. ARF can impact various organs, including the brain, heart, and lungs. AKI frequently occurs in hospitalized patients, particularly those in intensive care units and older adults. ARF is observed in up to 7 % of hospital admissions and 30 % of ICU admissions. Nonetheless, the condition is also prevalent among patients who are not critically ill [7]. AKI generally leads to gradual kidney function deterioration or ongoing dysfunction resulting in the irreversible loss of kidney cells and nephrons, potentially progressing to chronic kidney disease (CKD) [7] [8].

5.2 Chronic Kidney Disease (CKD) Overview

Chronic Kidney Disease (CKD) is a progressive disease of growing prevalence. In Europe, 100 million people suffer from CKD, making it the fifth leading cause of death globally by 2040. Additionally, CKD is among the most expensive diseases for health systems, with an estimated cost of EUR 140 billion annually in Europe [9].

5.3 Current Diagnostic Methods

Currently, creatinine is most diagnosed from the blood by the so-called Jaffé method or, more specifically, using enzymes [10]. These methods are one-time POC or laboratory tests. However, continuous monitoring could offer a more precise understanding of kidney function over time, minimizing the variability linked with standard periodic tests. This approach could enhance the management of patients' treatment plans and potentially improve outcomes by enabling immediate adjustments based on real-time data. Furthermore, it could provide a basis for more timely interventions for patients with chronic kidney disease (CKD) or acute kidney injury (AKI) [11].

Diagnostic imaging and biopsy are also used to assess kidney damage. Ultrasound, MRI, or CT scans can provide detailed images of kidney structure and function, though these scans can cost hundreds of euros per test and prices vary widely based on region, country, and health insurance. Kidney biopsy, where a small sample of kidney tissue is examined under a microscope, is another method. This procedure is often painful and can pose risks such as infection and bleeding from the biopsy site, with results taking more than 24 hours to obtain [12].

5.4 CKD Staging and Costs

CKD is classified into five stages based on the estimated glomerular filtration rate (eGFR). Stage 1 (G1) corresponds to eGFR > 90 ml/min and shows signs of kidney damage, Stage 5 (G5) refers to eGFR < 15 ml/min, indicating near-total loss of kidney function. The eGFR can be calculated using variations of the Schwartz equation, which incorporates serum creatinine results, patient age, gender, and height. This provides a measure of how many millilitres of waste the kidneys should filter per minute. In addition to blood tests for eGFR, urine tests for albumin and creatinine may be performed to determine the albumin ratio (ACR). Both for eGFR and ACR higher stages indicate increased severity of kidney disease.

The mean annual costs per patient vary considerably based on the CKD stage and treatment method, with estimates of [13]:

- **Stage 3a:** $3060, treated by medications to support blood formation, drugs for high blood pressure, vitamin D, medications for affecting phosphorus binders, and others. No drugs can affect creatinine levels.
- **Haemodialysis (stage 4 & 5):** $57,334, where the patient undergoes treatment around 3 times per week for 4 to 5 hours each. During this time, the patient's blood is filtered of small waste products through a membrane or filter and the creatinine levels return to normal.
- **Peritoneal dialysis (stage 4 & 5):** $49,490, differs from haemodialysis in a way that it is ongoing (daily) and can be undergone at the patient's home. It collects blood by washing the empty space

in the abdomen (peritoneal cavity). For some patients, this can be a significant setback, but it has one major advantage, which is not so fluctuating creatinine levels.

- **Kidney transplant (KT):** the initial cost of KT is $75,324 and followed up by subsequent tests and medications of $16,672, reaching $91,996 in total.

Kidney transplants (KTs) reduce the expected healthcare costs over 10 years by 66–79 % per patient on dialysis with the need for re-transplantation after 10-15 years [14]. However, the patient soon after KT can live a fulfilling life just like a person with normal, healthy kidneys.

5.4.1 Economic Burden and Projections

CKD clearly carries a significant economic burden that increases with disease progression and poses a significant risk and economic costs of other health complications. From 2021 to 2031, the prevalence of CKD is projected to rise by 10.5 % [15], more in **Appendix 1**. To mitigate the economic burden, cost-effective interventions targeting primary prevention and disease progression are essential. This has been validated through our meetings with CKD, ARF, and KT experts **Appendix 2**. Therefore, for CKD patients, we focus on monitoring creatinine levels and eGFR through the Schwartz formula, integrated into an app. This aids in preventing disease progression, optimizing dialysis treatment, and enhancing or maintaining patients' quality of life. It also assists doctors in deciding whether further diagnostic tests, such as ultrasound, CT, or MRI scans, are necessary.

5.5 Acute Renal Failure (ARF) Staging and Costs

Acute Renal Failure (ARF), also known as Acute Kidney Injury (AKI), is defined by:

- An increase in serum creatinine by 0.3 mg/dL or more within 48 hours.
- An increase in serum creatinine to 1.5 times or more of baseline within the prior seven days.
- Urine volume less than 0.5 mL/kg per hour for at least 6 hours.

ARF falls under Acute Kidney Diseases and Disorders (AKD), leading to irreversible kidney cell and nephron loss, potentially progressing to CKD. Therefore, our prototype focuses on the first two metrics for determining ARF and its severity, by observing the patient's kidney function trends.

5.5.1 Epidemiology and Economic Burden

As for the epidemiology overview, epidemiologists estimate approximately 18.5 million incident cases of ARF in 2023, forecasted to increase to 20.3 million by 2028, a 9.7 % increase in incidence [7] (**Appendix 3**). ARF can have an impact on various organs, including the brain, heart, and lungs. It frequently occurs in hospitalized patients, particularly those in intensive care units (ICUs) and older adults. ARF is observed in up to 7 % of hospital admissions and 30 % of ICU admissions. Nonetheless, the condition is also prevalent among patients who are not critically ill [16]. In the US, ARF affected an estimated 498,000 patients in 2019, with its annual cost ranging from $4.7 to $24.0 billion [17]. The most expensive patients were those that required dialysis. Given the sudden and unpredictable onset of ARF, preventing it is very unlikely. Therefore, our focus is on monitoring patients after AFR onset. Particularly on early diagnosis and severity assessment to implement cost-effective treatments and determine the need for dialysis, ultimately reducing expenses on extensive dialysis treatments by monitoring patient’s kidney function in response to current medications and interventions.

6. Business Feasibility

6.1 Business Model Canvas (Appendix 4)

6.1.1 Customer Segments and Market Potential

As for the identified customer segments, see customer segments analysis (**Appendix 5**). The need for continuous creatinine and kidney functions levels measuring devices is considerable. Mainly due to better sampling convenience, financial savings on other expensive diagnostic methods, reducing the workloads of ERs, nephrologists, nurses, and preserving time due to rapid and dynamic screening, where diet, activity, and other factors can influence the accuracy of one-time tests. Moreover, early interception, developing more effective treatment plans, and assessing the severity of kidney disease is important in reducing the growing economic burden and reducing the future incidence of kidney diseases. Details of the impact our device could have on current clinical practices for patients are in **Appendix 6**.

Currently, only Abbott Laboratories has a similar patented concept, but it requires further development. For more details refer to competition analysis (**Appendix 7**). Achieving TRL Stage 4 in July 2024 confirms our progress in detecting and quantifying creatinine levels (**Appendix 8**). Market analysis of how much revenue such device could generate annually in the Czech Republic is in **Appendix 9**.

6.1.2 Value Proposition

Our value proposition focuses on delivering continuous real-time monitoring of creatinine levels, which supports the early detection and management of chronic kidney disease (CKD) and acute kidney injury (AKI). This approach aims to alleviate the growing economic burden associated with costly medical treatments. The device will feature a user-friendly interface and integrate seamlessly with a mobile app or external hardware, ensuring accessibility and convenience. It is designed to be cost-effective compared to traditional diagnostic methods and will incorporate multiplex bioassay capabilities in the future to assess multiple biomarkers simultaneously. By reducing the workload for healthcare professionals, the device streamlines their processes and enhances patient outcomes through timely interventions. Additionally, it improves patient engagement and compliance, with potential integration with diet and exercise tracking for a comprehensive health management approach. Furthermore, by easing the workload and pressure on nurses, the device enables more personalized care, allowing nurses to spend more time with each patient and improve the overall quality of care.

6.1.3 Biomarker and Technological Focus

We focus on creatinine as it is essential for diagnosing kidney disease. Our device calculates eGFR using the Schwartz formula. Future iterations will include additional biomarkers like Cystatin C and NGAL for earlier and more accurate diagnosis. For more details regarding device iterations see **Appendix 10**.

6.1.4 Customer and User Engagement Channels

Our activities will include direct collaboration with healthcare professionals, who will be incentivized with commissions or other rewards for recommending our biosensors. This will also involve regular meetings to gather feedback on the device, discuss future development plans, and publish articles. Additionally, we will raise awareness about kidney-related issues and the benefits of our biosensors by participating in conferences and showcases, such as the ERA Congress and MEDICA in Düsseldorf, as well as through lectures and workshops. To boost public visibility, we plan to engage celebrities like Mr. Ivo Šmoldas, a well-known Czech TV and radio presenter and actor (**Appendix 11**). To better address user needs and facilitate further development, regular workshops will be held. Additionally, a customer service department will be established to quickly resolve any issues encountered by users.

6.1.5 Sources of Income

Initially, we will sell the device to hospitals and health centers for €109 each. A mobile app-connectable version will later be available for €89. The device is expected to operate for one week.

6.1.6 Key Activities

Post-R&D, we will focus on market engagement and finalizing technical and regulatory groundwork.

Market Strategy

We aim for CE mark compliance ensuring compliance with relevant legislative standards. We aim to develop user-friendly application compatible with mobile devices. Currently, the app design is in its preliminary stage (**Appendix 12**). Future iterations aim to incorporate diet and exercise tracking.

Our marketing team will secure storage, delivery partners, and investors. Market positioning strategy includes promoting brand visibility, developing an expansion strategy and maintaining strong relationships with our end customers and business partners.

Regulatory and Technical Development

Initially, we will file for a patent through UCT's DTT. Based on pre-existing agreements, the authors will form an LLC and acquire a patent license from UCT, which will be free for the first two years. Securing sufficient funding to enter preclinical and subsequent clinical trials through seed funding from grants, accelerators, and venture capitalists such as the i&i Biotech Fund. Quality assurance will be conducted following European Medicines Agency (EMA) standards, aiming for a CE mark. This process will include meeting all legislative requirements necessary for market entry and manufacturing.

Strategic Advisory and Expansion

As we approach the end of the R&D phase, we will establish a board of advisors to navigate unforeseen challenges and bolster the company's credibility. Potential advisors include doc. Ing. Lenka Švecová, Ph.D. (startup transfer expert), Ing. Jaromír Zahrádka, Ph.D. (CEO of i&i Biotech Fund), and doc. Ing. Zdeněk Slouka, Ph.D. (current team supervisor), who have already provided mentorship throughout this project.

6.1.7 Key Resources

Our multi-disciplinary team includes experts in chemical and bioengineering, biochemistry, sensorics and cybernetics, nano and microtechnologies, and economics. We also collaborate with nephrology and veterinary experts and i&i Biotech Fund for business aspects.

Over the next three years, our priority is advancing the prototype through research at UCT Prague, supported by UCT's Department of Technological Transfer. For market launch, we aim to expand production partnerships. Currently, potentiostats are assembled in-house, chemicals from Merck, and enzymes from Alchimica and Merck. Screen printed electrodes are sourced via UPCE. Initial R&D funding comes from UCT Prague, with additional funding sought from i&i Biotech Fund's incubation program and external grants. For further details regarding project timeline see **Appendix 13**. For better understanding what should be our focus, SWOT analysis (**Appendix 14**) and MOSCOW analysis (**Appendix 15**) have been carried out.

6.1.8 Key Partners

Our key partners include hospitals/health centres, insurance companies, pharmacies, and e-pharmacies. Future collaborations will extend to veterinary clinics, sports organizations, retirement/care centres, and palliative care providers.

During the R&D phase, we will work with UPCE on SPE optimization and semi-industrial manufacturing, producing up to 100,000 SPEs daily. Scale-up options will be explored with UPCE and BVT Technologies. We will secure deals with suppliers like BBI Solutions and Creative Enzymes and consider biotechnological enzyme production. For initial funding and commercialization guidance, we will collaborate closely with the i&i Biotech Fund.

6.1.9 Financial Viability

Manufacturing and Cost Analysis

The manufacturing costs are estimated at €78.5 for large scale manufacturing. The price is highly influenced by enzymes cost. Therefore, to reduce the device price we rely on enzyme regeneration methods.

Establishment of a long-term manufacturing facility, streamlining our distribution network and securing contracts with production partners is crucial for ensuring financial sustainability and maintaining market competitiveness.

Pricing Strategy

Hospitals, healthcare centres, dialysis centres, and other relevant entities can purchase the complete diagnostic package for €109. Future devices connected to mobile phone will be available for €89. A detailed device cost analysis is available in **Appendix 16**.

Research and Development Costs

The projected expenses for additional research, development, and device optimization amount to €150,000, to be fully covered by UCT Prague. This strategy aligns with the decision to transfer patent rights to UCT Prague. Minimizing external partnerships is deemed essential for the company's establishment.

Patenting and Licensing Expenses

Initial costs for obtaining patent and licensing rights are estimated at €15,000 over the first two years, covered by the University of Chemistry and Technology, Prague. Subsequent annual costs, including patent rights rental and maintenance fees, are projected at €4,000, to be borne by the company.

Pre-clinical Testing Costs

Pre-clinical testing is estimated to cost €132,000, covering all aspects of testing in compliance with GLP standards. The studies will evaluate cytotoxicity, genotoxicity, and other factors on rats, rabbits, in vitro tests, and bacterial analysis. The study duration will adhere to multiple ISO (e.g. 10993-1 (2021)) protocols. Specific details remain confidential as per the final offer from the preclinical studies company.

Clinical Trials and CE Marking

For clinical trials, the projected cost is €2,540,000. This estimation includes expenses for staff, laboratory rental, evaluation and others (**Appendix 17 & 18**). To obtain CE marking for the device, comprehensive risk assessment and the creation of an instruction manual will be addressed. Details in **Appendix 19**.

Early Market Launch and Facility Costs

Annual expenses for laboratory and office rentals are approximately €33,000 and €46,368, respectively. For more details refer to **Appendix 20**.

Market Penetration Strategy

To penetrate the market, we will establish commission-based agreements with doctors, encouraging them to prescribe our product and generate word-of-mouth referrals. We plan to expand distribution channels through partnerships with influential figures and organizations in kidney health. Our marketing strategy will focus on a product-led approach, gathering feedback from satisfied customers through in-person meetings and structured questionnaires. This data will inform improvements to user manuals, onboarding processes, UX/UI interface, and marketing campaigns, optimizing our product based on user preferences. For more details, refer to **Appendix 21** or [18].

For health insurance companies, we aim to collect and analyse data demonstrating the cost-effectiveness of our device, highlighting its potential to reduce the economic burden of kidney treatment. To prepare for market launch FMEA draft has been carried out (**Appendix 22**). This serves as a living document and will be periodically updated.

Sales and Expansion Strategy

Initially, we will sell the device to hospitals and health centres with external monitoring hardware. Post-launch, we will optimize for a sustainable market position before expanding to mobile-connected devices. Each device's price is adjusted by roughly 35% of the manufacturing price. To break even, 31345 devices must be sold annually (**Appendix 23**). Expansion plans include entering the German and Nordic markets for better penetration and higher prices. For comprehensive financial analysis, refer to **Appendix 23 and 24**.

7. Team and Support

7.1 Contribution of the Team Members

During biosensor development, the team was initially divided into three laboratory sub-teams, which aimed to evaluate the feasibility of the three chosen detection principles further. The chosen detection principles included enzymatic cascade combined with electrochemical detection, aptamers and MIPs (molecularly imprinted polymers) combined with optical detection. Later, the sub-teams were merged into two sub-teams focusing on detection (flow cell design and physical transduction) and enzymatic cascade.

***Table 1**: Contribution of each team member.*

Member	Contribution
Adam Benjamin Plšek	**Explored preparation of MIPs and physical** transduction (namely molecular transport and electrochemical setup),
Daniel Křížek	**Co-captain; founder; head of the translation potential team**; secured multiple sponsors, collaborations, and initial funding from UCT Prague; responsible for finding mentors, experts, labs, team members, and suppliers; explored preparation of MIPs and physical transduction; coordinated meetings and sub-teams' cooperation and progress
Denis Najman	**Worked on physical transduction**, participated in communication with in-kind sponsors and experts
Eva Vogelová	**Co-captain**; participated in the recognition element development (aptamers and enzymes); facilitated organization of lectures and cooperation with FZU; member of the social media sub-team
Hynek Housar	**Member of the translation potential sub-team**, worked on securing sponsorships and collaborations, explored the overall feasibility of our biosensor, worked on physical transduction
Ivana Venkrbcová	**Explored biorecognition elements**, worked on optimization of the electrochemical set-up and ensured accurate and reliable measurements through fine-tuning
Ján Cúth	**Worked on response time validation using COMSOL modelling**, contributed to the translation potential team, prepared materials, conducted nephrology expert and business interviews, wrote the team results document
Jan Šťovíček	**Head of the physical transduction team;** methodology and evaluation of electrochemical measurements, development/optimization of a sensitive electrode for the end analyte, consultation on sensor design, conceptualization of the preparation of MIPs
Jan Vyčítal	**Designed, manufactured and tested potentiostat**, supervised mobile app development with an accessible user experience that would gather, store, and present measured data
Jiří Kohout	**Participated in the recognition element development** (aptamers and enzymes), theoretical research, crafting of marketing materials and social media content, and communication with external distributors
Marek Plachý	**Responsible for mobile app development** with accessible user experience that would gather, store, and present measured data; worked on hardware testing; worked on securing sponsorship with SOTIO
Tadeáš Sochor	**Participated in the recognition element development** (aptamers and enzymes), member of the social media sub-team, administered our LinkedIn profile, and created several posts and captions, facilitated connections with external distributors and affiliated laboratories at UCT Prague
Terézia Horníková	**Participated in the recognition element development** (aptamers and enzymes), member of the social media sub-team, facilitated contact with an expert on electrochemical detection

7.2 People Who Have Given Support

We would like to thank doc. Zdeněk Slouka, Ph.D. who provided us with guidance and support throughout the competition as our mentor. We would also like to thank Dominik Králík, doc. Oleksiy Lyutakov, Barbora Holubová, Ph.D., Ludmila Karamonová, Ph.D., Elena Miliutina, Ph.D., Roman Elashnikov, Ph.D., doc. Lenka Švecová, Ph.D., doc. Hana Scholleová, Jaromír Zahrádka, Ph.D., Ph.D., RNDr. Hana Lísalová, PhD. and Juditha Anthi, Ph.D., for their support at some point on our journey through providing insights, trainings or feedback. We are grateful to medical/veterinary experts, namely Ivan Šebesta, CSc. and Simona Kovaříková, Ph.D. for providing key insights into the given topic. Moreover, we would like to thank Kateřina Kovaříčková from DTT for her help in establishing the team and her administrative support. Lastly, we would like to thank all companies and individuals who provided us with finances, materials, or feedback on our ideas. Lastly, we thank Ivo Šmoldas, CSc. for sharing his patient journey with us.

7.3 Sponsors

UCTeam would like to extend our heartfelt gratitude to our partners and sponsors for their time, comprehensive feedback, guidance, financial support, and more. Their contributions have been instrumental in enabling our participation in the competition and beyond.

***Table 2**: List of sponsors and partners.*

Sponsors	Type of support
UCT Prague	financial support, in-kind contribution (laboratory provision, technical advice)
FZU (Czech academy of life sciences)	in-kind contribution (laboratory provision)
BVT Technologies	in-kind contribution (potentiostat and related hardware, SPEs)
SOTIO Biotech	financial support
Mars Wrigley	financial support
Parker Hannifin	financial support
Henkel	in-kind contribution (technical advice - conductive inks)
UPCE (University of Pardubice)	in-kind contribution (SPEs, conductive inks)
MediTox	in-kind contribution (technical advice)

8. Final Remarks

During the development of our biosensor, our team initially divided into three lab sub-teams to evaluate the feasibility of three detection principles: enzymatic cascade with electrochemical detection, aptamers, and MIPs (molecularly imprinted polymers) with optical detection. As the project evolved, we reorganized into two main groups—focusing on detection (flow cell design and physical transduction) and enzyme immobilization—while also forming a team dedicated to reader instrument and app design. Two additional sub-teams handled marketing (PR, social media) and the project's translation potential. Each sub-team had a sub-leader responsible for tracking progress and setting goals, while overall leadership was shared by two co-captains.

Despite challenges, particularly supplier delays, our experience in this competition has been invaluable. We've grown as researchers and future professionals, gaining new skills and insights. The KidneyGuard prototype we developed offers significant value to patients and the public. We hope for its continued development, optimization, and eventual market launch after meeting all legislative standards. Additionally, as the first Czech team to compete, we hope to inspire future UCT Prague students to participate.

The UCTeam extends heartfelt gratitude to everyone who supported us through interviews, meetings, and guidance. Your help has been invaluable. We are also thankful for the financial support, materials, devices, and research spaces provided, which were crucial to our participation.

Special thanks to our supervisor, doc. Ing. Zdeněk Slouka, Ph.D., for his unwavering guidance. We also appreciate the invaluable feedback from doc. Ing. Lenka Švecová, Ph.D., and doc. RNDr. Ing. Hana Scholleová, Ph.D., on the economic, translational, and business aspects of our project.

Finally, we thank our sponsors for their generous contributions and proudly acknowledge their support, showcasing their logos on our apparel during the SensUs Innovation Days 2024 in Eindhoven, Netherlands.

Appendix

Appendix 1: Chronic Kidney Disease: Epidemiology Forecast to 2031

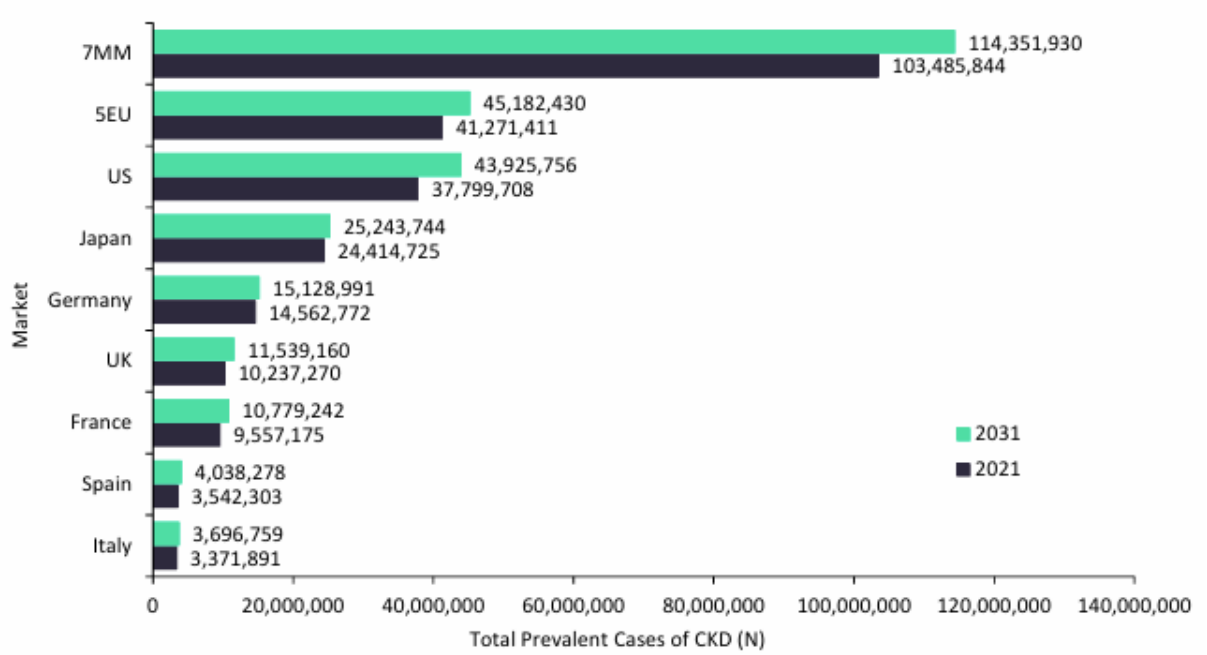


***Figure 4**: Total prevalent cases of CKD, both sexes, N, ages ≥ 18 years, 2021 and 2031 (7MM: US, Japan, Germany, UK, France, Spain, Italy) [15]. Source: GlobalData; (Imai et al., 2009; Otero et al., 2010; Bongard et al., 2012; Jameson et al., 2014; Nakai et al., 2014; De Nicola et al., 2015; Girndt et al., 2016; Centers for Disease Control and*

Appendix 2: Interviews Summary

Veterinary experts - **MVDr. Simona Kovaříková, PhD.** - veterinary expert - internal medicine of cats and dogs, professor at the University of Veterinary Sciences Brno with various publications regarding kidney problems of cats and dogs; **MVDr. Jaroslav Kučera, CSc.** - a private veterinary specialist with multiple publications regarding kidney problems of cats and dogs; Currently the most common method for creatinine detection for veterinary purposes is the Jaffé method. The total cost of one sample is roughly 15 euros. The cost of full kidney profile blood testing for one sample is roughly 40 euros (labor not included). This method of detection is not ideal as it often strongly varies in between different laboratories (big differences in reference ranges). Also, in this regard, the critical levels of creatinine are very patient specific, as its breed, weight, muscle mass, age etc. come to the equation. KidneyGuard can help track creatinine trends of veterinary patients, which can potentially help veterinary experts gain broader understanding of kidney related problems regarding those criteria. Secondly, as a lot of commonly used drugs (cytostatics), analgesics (mainly nonsteroid antiphlogistic) or surgeries (e.g. interventions after sepsis or purulent inflammation of the uterus) done of different organs may cause nephrological damage, it is desirable to monitor patients especially pre- and post-surgery and while medicated with potential side effects. Monitoring of creatinine trends is also desirable when treating/monitoring nephrological patients of small weight, especially below 10 kilograms. For those patients, repetitive blood sampling is very invasive, therefore they would benefit from less invasive monitoring methods. For patients suffering from CKD, the device will most likely not be desirable. Now CKD patients are mostly monitored quarterly while already on a specific diet. To make our device more viable for the patients ówners. Our device ought to detect more biomarkers so that additional visits are saved. Detection of other markers such as SDMA or FGF23 could serve this purpose. Multiplexing ability of course would be beneficial in all cases of veterinary monitoring.

MVDr. Zuzana Drábková, Ph.D., Dipl. ECEIM - veterinary expert - internal medicine of horses, professor at the University of Veterinary Sciences Brno; As horses or any other big animals do not suffer from kidney malfunctions nearly as much as cats or dogs (neither AKI nor CKD), continuous monitoring might only be

interesting for research purposes of veterinary experts or when the patients are subjected to a surgery and/or treatment which might be potentially harmful to their kidneys (similarly to cats or dogs).

Medical experts - **MUDr. Ivan Šebesta, CSc.** - nephrologist, professor at Charles University; **Doc. MUDr. Květa Bláhová, CSc.** - paediatrician with specialization in internal medicine and nephrology, professor at Charles University - Much like for the veterinary patients, our biosensor would most likely not be necessary for patients suffering from CKD. However, being able to track trends in patients' creatinine levels would be beneficial for patients pre and post surgeries (including kidney transplant), patients medicated with potentially problematic kidney response and patients with ongoing dialysis. Secondly, groups such as oncology patients or gene therapy patients can benefit from KidneyGuard as monitoring kidney function while undergoing treatment is of important role due to kidneys being the first organs impacted by harmful side-effects potentially resulting in kidney malfunction. Thirdly, a potential market for high performance athletes and sport organizations was discussed. In this context, KidneyGuard can be used when optimizing the training plan for athletes as well as preventing rhabdomyolysis. NGAL and Cystatin C were also discussed as biomarkers that can help unravel the state of patients' kidney health.

Appendix 3: Incidence Cases of ARF in 2023 and 2028

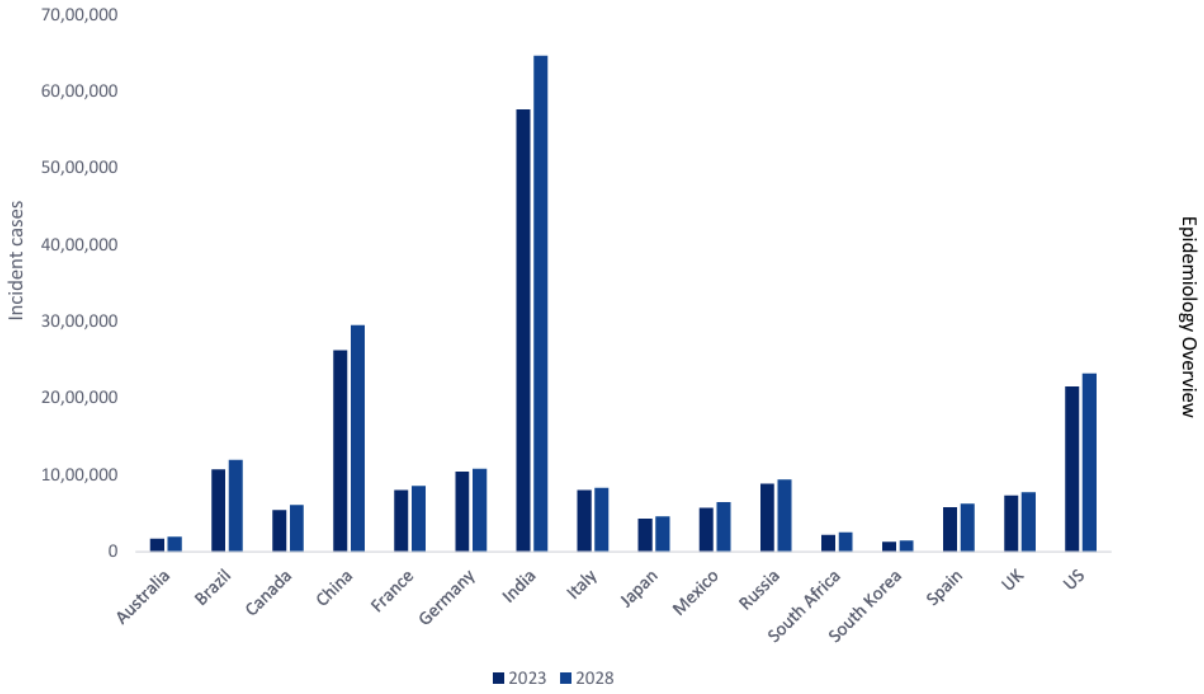


***Figure 5**: Incidence cases of ARF in 2023 and 2028 in the 16 major pharmaceutical markets (16MM: Australia, Brazil, Canada, China, France, Germany, India, Italy, Japan, Mexico, Russia, South Africa, South Korea, Spain, the UK, and the US) [7]. Source: Source: GlobalData, Pharma Intelligence Center, Epidemiology & Market Size Database. Based on peer-reviewed literature, disease registries, and primary research. Note: Cases represent 2023 and 2028 incident cases alleges (>=20 Years).*

Appendix 4: Business Model Canvas

***Figure 6**: Business Model Canvas.*

Appendix 5: Customer Segments Analysis

***Table 3**: Evaluation of customer segments.*

Customer Segment	Compelling Reason to Buy	Implementation Difficulties	Other External Risks
Nephrology & Dialysis Centres	Monitors creatinine and eGFR for CKD and ARF, aiding in personalized, cost-effective treatments.	Desire for additional monitoring (potassium, sodium, Cystatin C) complicates development.	Challenges include commission agreements and competition from companies like Abbott Laboratories.
Oncology & Gene Therapy Centres	Detects kidney issues early, enabling safer treatment plans and significant financial savings.	Small market size: one-time creatinine tests might be a more cost-effective alternative.	Competitors offer cheaper one-time tests.
Intensive Care Units (ICUs)	Easy to operate, quick results via app, reduces nurse workload and improves patient care.	Concerns about ease of connection and simple UX/UI; reliability and app functionality are crucial.	Ensuring reliability; potential need for an external server and future storage infrastructure.
General Practitioners (GPs)	Assists in monitoring kidney function, managing referrals, and reducing workload.	Convincing GPs to adopt commission-based agreements; one-time tests often more cost-effective.	One-time testing is usually sufficient and cost-effective for initial assessments.
Veterinary Clinics	Monitors kidney function in animals during and after treatment, crucial for early intervention and lifespan extension.	Developing a small, accurate device that is comfortable for animals is challenging.	Keeping the device affordable to maintain interest; cost is not fully covered by users.
Health Insurance Companies	Early intervention and personalized care reduce expenses and economic burden.	Establishing contracts is challenging; compelling data needed to demonstrate cost savings.	Demonstrating superior cost-effectiveness against competitors' strategies.
Pharmacies & E-Pharmacies	Sales commissions for devices purchased with prescriptions incentivize promotion.	Device reliability is crucial; establishing contracts and setting up a customer service department for malfunctions.	Competitors may offer more cost-effective solutions; need to stay informed and address issues promptly.
Sports Doctors and Coaches	Optimizes training plans by tracking kidney function and how it relates to performance.	Device must be appealing for regular use; currently seen as a one-time purchase per season.	Assessing the correlation between kidney function, diet, activity, and performance is essential.
Retirement Homes & Care Centres	Provides valuable kidney health insights for elderly patients on multiple medications.	Demonstrating that ongoing benefits justify the cost compared to cheaper one-time tests.	Competitors offer cost-effective one-time tests, impacting market adoption.

When exploring the market and evaluating where our biosensor could be of interest, we identified four key customer segments:

1. Hospitals and health centres, consisting of nephrology centres, dialysis centres, Intensive Care Units (ICUs), ERs, doctors and general practitioners, and oncology and gene therapy centres.
2. Health insurance companies.
3. Pharmacies.
4. Veterinary clinics (future).

Appendix 6: Patient Journey and UCTeam KidneyGuard Impact Overview

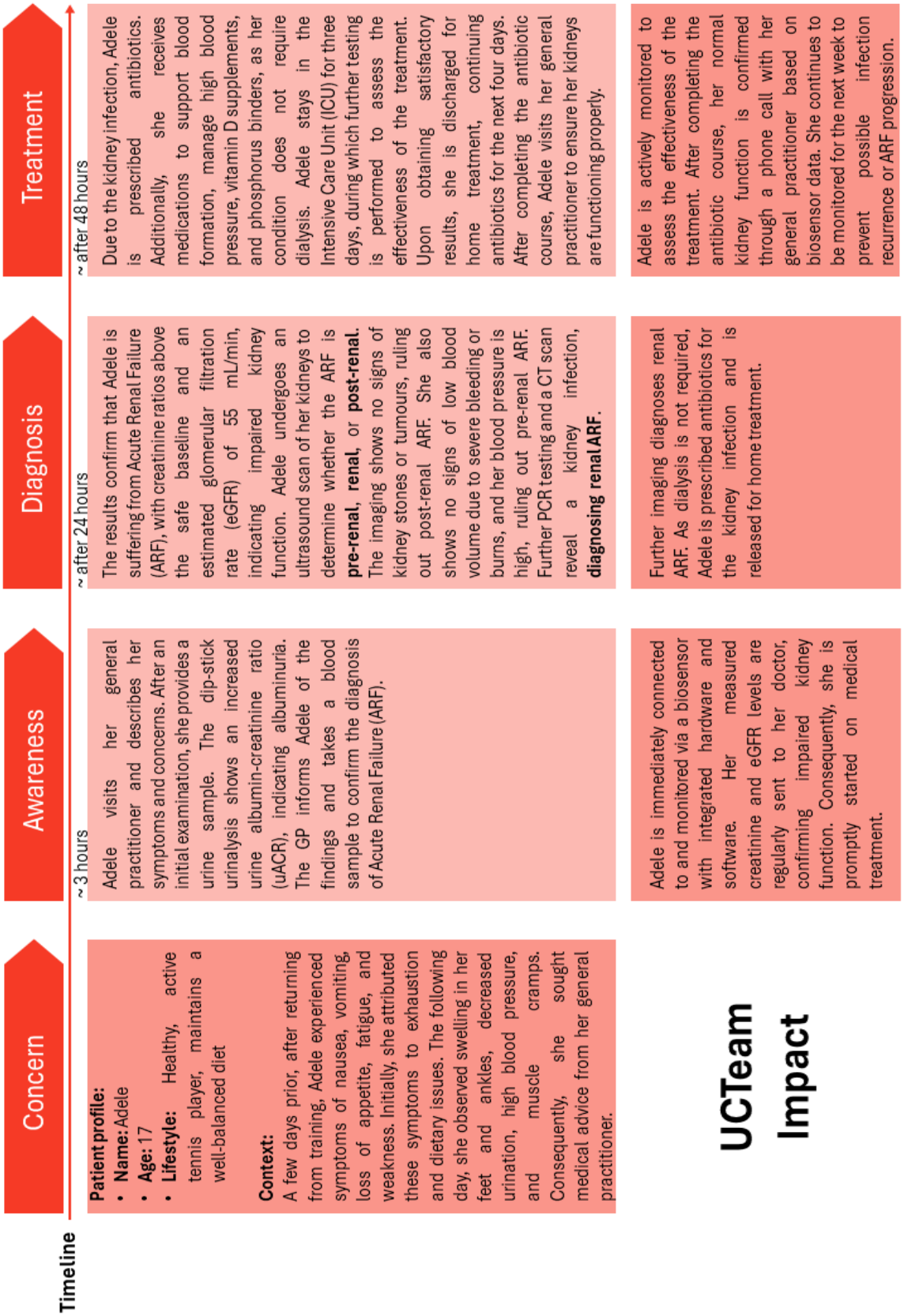


***Figure 7**: Patient journey and UCTeam KidneyGuard impact overview on current clinical procedures.*

Appendix 7: Competition Analysis

***Table 4**: Competition analysis.*

Established diagnostic methods					
Method	**Advantage**	**Disadvantage**	**Pricing**		
Jaffé method [19]	low cost	nonspecific reactions, susceptibility to interferents, interlaboratory variability [20]	commercially available kits ~ 0,35 € per sample (reagents), ~14 € in hospitals including the cost of blood withdrawal*		
IDMS	high specificity, good reference method	high cost	> 200 000 € - HPLC-MS/LC-MS machinery [21]		
HPLC	high specificity, good reference method	high cost	> 50 000 € - HPLC machinery [21]		
Enzymatic method [19]	high specificity	relatively high cost	~ 2 € per sample (reagents), > 2500 € - spectrophotometer [22]		
Competitors					
Type	**Range**	**Sample size**	**Note**	**Pricing**	**Matrix**
Abbott i-STAT Creatinine Test Cartridge	18-1768 µmol/L	65 µL		200 - 350 €/25 samples	blood
Nova Maxx Nova Max Creatinine/eGFR	27-619 µmol/L	blood drop (1,2 µL volume of the strip)			blood
Nova Maxx StatSensor Creatinine Connectivity Meter	27-1056 µmol/L	blood drop (1,2 µL volume of the strip)			blood
Afinion™ 2	multiplex device suitable for laboratories, provides ACR (suitable for diabetes patients) – spectrophotometry				blood
Afinion™ ACR	provides ACR (suitable for diabetes patients) - spectrophotometry				urine
Competing patents					
Abbott - US2020/0241015 A1	the most similar patent to our current technological setup				
KidneyGuard UCTeam					
Method	**Advantage**			**Disadvantage**	**Pricing**
Enzymatic method	high specificity, continuous readings, dynamic values (influenced by diet & physical activity), generic/adaptable concept (multiple biomarkers in the future), connected to user-friendly application/external hardware, easy to control/connect and use even by untrained people			not ready for market launch (in development), higher environmental burden than one-time strips	€89/device for 7 days, €109/device with reader for 7 days

* Rough prices given by the medical professionals or [19].

Appendix 8: Technology Readiness Level (TRL) Assessment

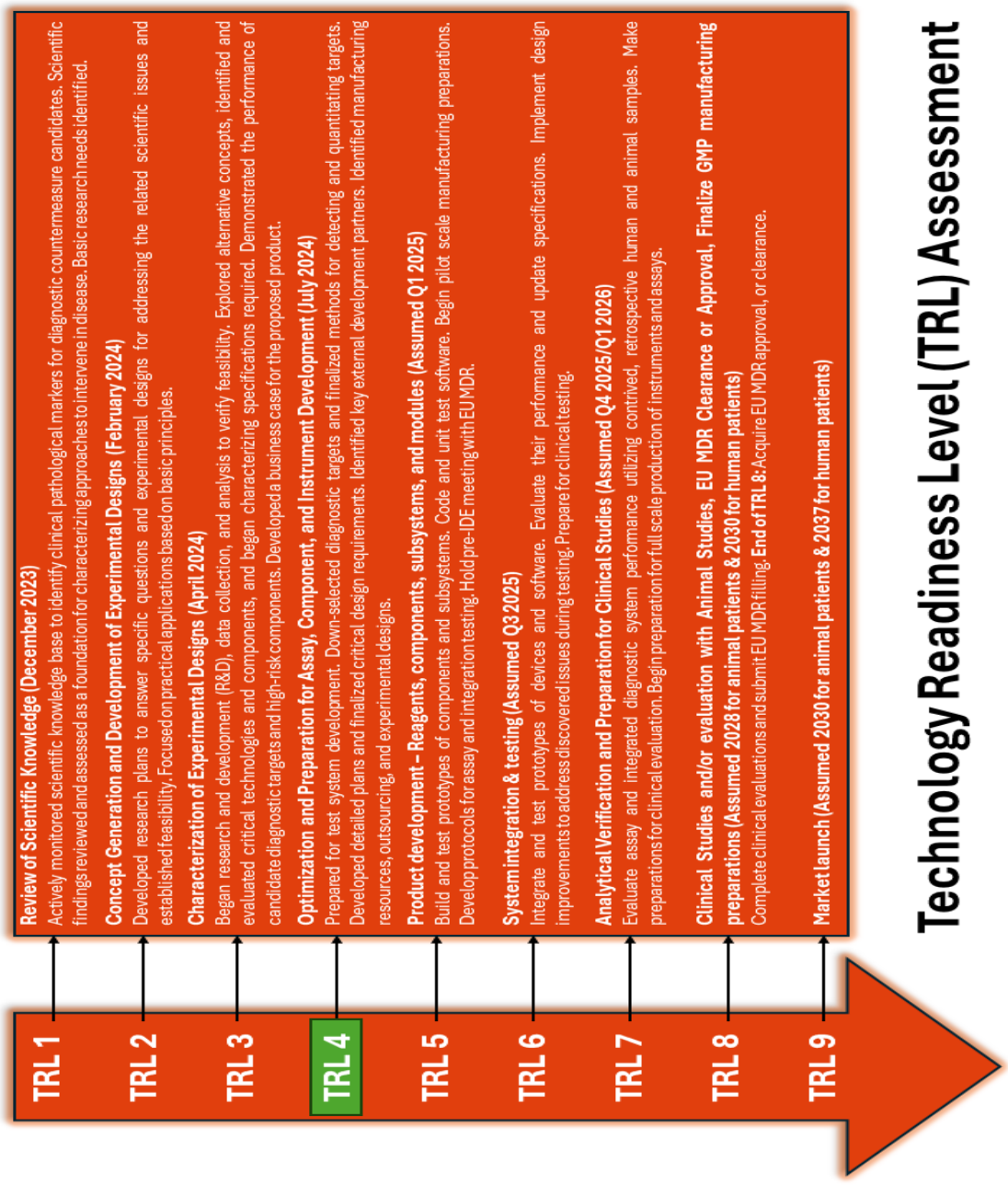


***Figure 8**: Technology readiness level assessment [23].*

Technological Readiness Level (TRL) Assessment

As of July 2024, we have completed TRL Stage 4, finalizing methods for detecting creatinine levels and setting design requirements for our biosensor. We are collaborating with development partners to secure manufacturing resources and prepare for in-lab production. Our goal is to progress through the TRL stages to reach TRL Stage 9.

Appendix 9: Market Analysis (TAM, SAM, SOM)

***Table 5**: TAM SAM SOM market analysis in the Czech Republic.*

Data input for market analysis calculation		
# of hospitalizations in CZ in 2016 [24]	2.37	million
CZ population	10.70	million
Global CKD prevalence [25]	9.50%	
Global AKI prevalence per hospitalizations [26]	10.00%	
Global cancer prevalence [27]	0.25%	
Global gene therapy patients' percentage	0.10%	
Expected annual revenue per patient	300	€
SAM - Serviceable Available Market -> 20 % of TAM		
SOM - Serviceable Obtainable Market -> 10 % of SAM		
TAM - Total Addressable Market (mil. € annually)		
CKD - Chronic Kidney Disease		304.95
AKI - Acute Kidney Injury		71.07
Oncology patients		8.03
Gene therapy patients		3.21
Total	**mil. €**	**387.26**
SAM - Serviceable Available Market (mil. € annually)		
CKD - Chronic Kidney Disease		60.99
AKI - Acute Kidney Injury		14.21
Oncology patients		1.61
Gene therapy patients		0.64
Total	**mil. €**	**77.45**
SOM - Serviceable Obtainable Market (mil. € annually)		
CKD - Chronic Kidney Disease		6.10
AKI - Acute Kidney Injury		1.42
Oncology patients		0.16
Gene therapy patients		0.06
Total	**mil. €**	**7.75**

As for the market analysis calculations, we can clearly see as identified through interviews, that it is in the best business interest to focus mainly on CKD and AKI patients, as oncology and gene therapy markets represent a small part of the overall market share. For the calculations, SAM represents 20 % of TAM, SOM represents at least for the early stages only 10 % of SAM.

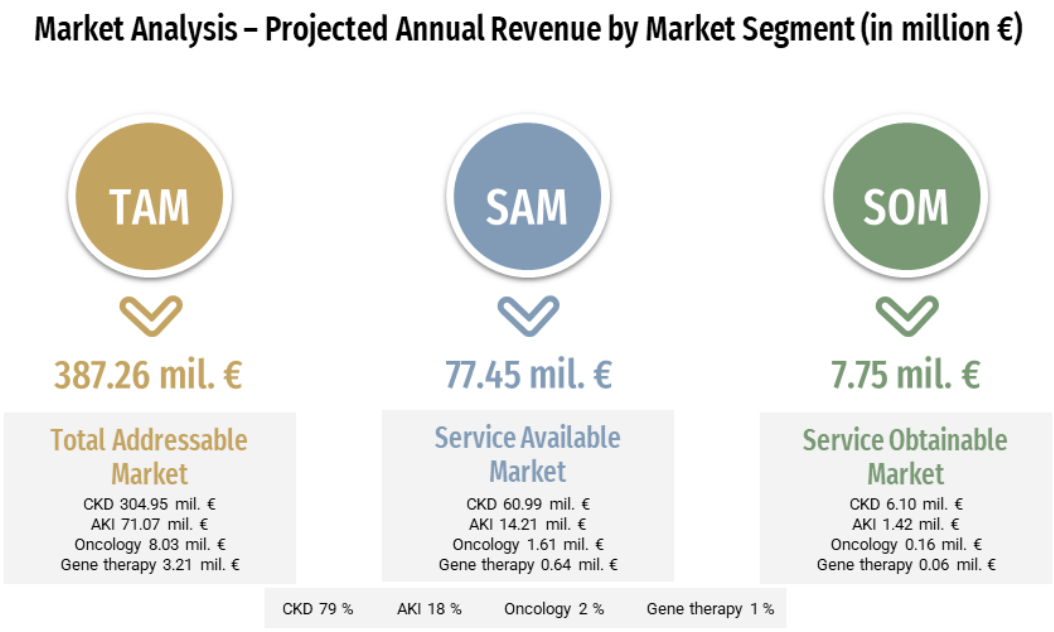


***Figure 9**: TAM SAM SOM market analysis graphical overview in the Czech Republic by market segment.*

Appendix 10: Device Iterations and Development Overview

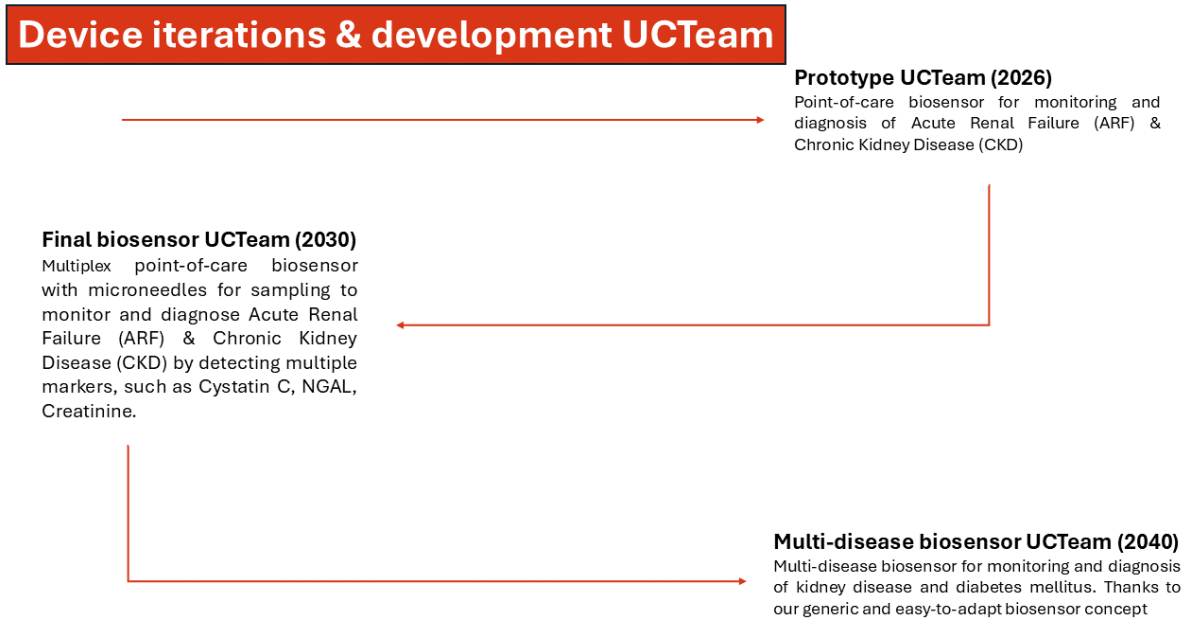


***Figure 10**: Device iterations and future device development plans.*

Device Iterations and Future Development Plans

Our initial product will be a biosensor utilizing electrochemical recognition based on an enzymatic cascade reaction. During this period, we plan to develop a multiplex point-of-care device to monitor multiple biomarkers, including Cystatin C, NGAL, and Creatinine, for better, earlier, and more accurate recognition of kidney issues. Looking ahead, our vision includes creating a biosensor capable of multi-disease monitoring, incorporating various biomarkers and enzymes to detect multiple conditions simultaneously, enhancing comprehensive health monitoring within a single device.

Appendix 11: Patient Interview, Ivo Šmoldas

Ivo Šmoldas is a famous Czech poet, translator, screenwriter, cultural publicist, and presenter. He suffers from polycystic kidney disease and shares his journey to a kidney transplant with a great sense of humour. As Ivo Šmoldas likes to say: „While everyone around me inherited wealth, I inherited diseases. “Polycystic kidney disease is a condition that mainly affects the kidneys, causing them to deteriorate as cysts accumulate and functional tissue diminishes. „In the initial stages, there was no treatment since there were no symptoms to manage. However, I eventually (aged 30) started receiving medication for high blood pressure, a secondary symptom of the disease caused by the loss of functional kidney tissue, leading to increased pressure in blood vessels. “Regular check-ups, including blood tests and urine analysis, became more frequent over the years. Ivo received a kidney transplant 3 years ago. It involved a thorough evaluation process to determine a patient's eligibility for a transplant and their overall health condition before being placed on the transplant list. The waiting time for a donor organ varies but generally ranges from one to one and a half years on average. He was lucky to get the transplant after waiting only 3 months. The transplant procedure is a major intervention, but the general anesthesia makes the process relatively painless. After being discharged from the hospital, he had to visit the hospital every day for check-ups. Initially weak and fatigued, it took some time for his body to adapt to the new kidney. Besides being fatigued, he also suffered from anemia because of the surgery. We discussed the idea of using a creatinine biosensor to monitor kidney function. From a patient's perspective, having a continuous monitoring system would not only enhance patient comfort but also offer a more convenient and efficient way to monitor health progress. „Continuous monitoring could ensure timely alerts for any deviations in health parameters, especially during the critical phases for pre- and post-transplant patients. It would be a great way to reduce the

number of check-ups. “The idea of having a remote monitoring system, possibly through online platforms, could streamline the follow-up process and provide peace of mind for both patients and their families, even years after the surgery.

Appendix 12: Current Application Design

The Creatinine biosensor measures real-time creatinine data, which we aimed to present in a user-friendly manner to the end user (patient/doctor) and store efficiently. While a laptop connected via USB cable served well for testing, it was not practical for real-world application. This led us to develop a stand-alone mobile application with a strong focus on user experience. To ensure its success, we sought direct feedback from patients, which guided our design decisions.

The communication between the measuring device and the mobile application is facilitated using Bluetooth 4.0 - Low Energy technology, which utilizes special profiles dedicated for wearable sensor data transfer. The application is primarily written in Kotlin, a language recommended by Google, and makes use of Jetpack Compose, a modern framework for GUI creation. The received data are securely saved in an SQLite database through the mediation of Room abstract layer, ensuring secure database access.

In the future, we would like to further iterate on the application, making it lighter and “user-friendly”-according to received feedback. Further improvements include extending support for iOS devices or/and developing a modern, web-only interface, which was initially discouraged because of the need for deep hardware interaction and related issues. Gained patient data could be relevant for research on kidney diseases or further biosensor development and, therefore, worth collecting. However, that would require a central backend database, which brings new technical and legal obstacles.

Appendix 13: Project Timeline

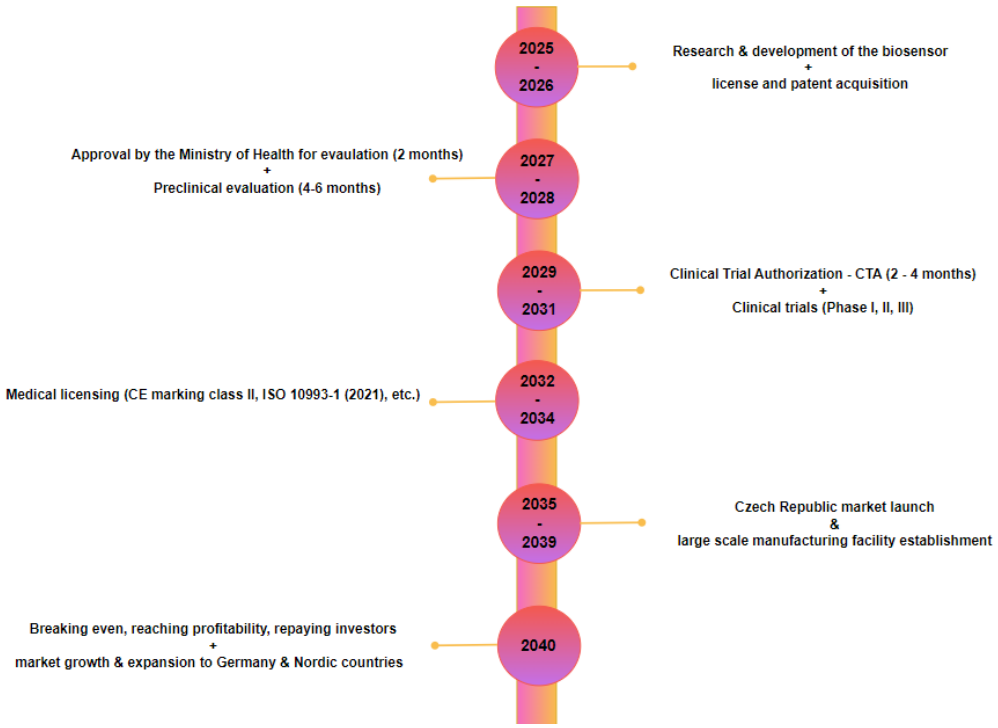


***Figure 11**: Project timeline and planning overview.*

Project Timeline and Planning for Biosensor Development and Market Introduction

Our startup aims to bring a biosensor for continuous creatinine monitoring to market within 10 years, with buffer time for potential setbacks. Here is a concise timeline of our activities:

Research & Development (2025-2026) We will finalize R&D over two years, secure a biosensor patent, and prepare for preclinical evaluation.

Preclinical Evaluation (2027-2028) We will request preclinical evaluation approval from the Czech Ministry of Health. Partnering with a GLP-certified organization, we will conduct tests on three non-rodent subjects over 4 to 6 months to ensure safety and reliability.

Clinical Trials (2029-2031) After successful preclinical tests, we will seek Clinical Trial Authorization (CTA). Clinical trials will proceed in four phases:

- **Phase I:** Test on 20 to 80 healthy volunteers for safety.
- **Phase II:** Test on 100 to 300 kidney disease patients for effectiveness and safety reassessment.
- **Phase III:** Large-scale testing on 1,000 to 3,000 kidney disease patients.
- **Phase IV:** Ongoing monitoring post-market launch [28].

Regulatory Approval and CE Marking (2032-2034) We will pursue EU regulatory approval and CE Marking, a process that may take up to two years.

Manufacturing and Market Launch (2035) By 2035, we plan to establish large-scale manufacturing and introduce the biosensor in the Czech Republic, expanding to Germany and Nordic countries based on initial success.

This timeline outlines our strategic approach to biosensor development, ensuring thorough research, regulatory compliance, and market readiness for a successful launch and expansion.

[29-31]

Appendix 14: SWOT Analysis

***Table 6**: SWOT analysis.*

Strengths: The team is very motivated and offers a variety of talents (hardware and software development, material chemistry, microfluidics, biochemistry, business, marketing, etc.). - The team has already established important partnerships that can help accelerate the R&D phase and the transition to the manufacturing phase. Moreover, some of the potential partners offer experience with biotechnological start-ups and a vast network of contacts. - The team's alma mater is keen to support the continuation of this project beyond the competition's final. That being said, UCT enables the team to continue the R&D phase with its full support and subsequently offers extremely favourable conditions for using patent licenses. - The team's established good relationships with research experts as well as people from the area of MedTech transfer, who are willing to take part in the board of advisors eventually. - The KidneyGuard has significant potential to be redesigned to detect multiple biomarkers. This enhancement would not only improve its accuracy but also expand its applicability to new customer groups.
Weaknesses: Compared to our current prototype, some of the approaches and components used already have patents. - The prototype still needs many adjustments within the R&D phase to function in a customer-friendly wearable manner and simply be market ready. - The team is relatively small and has limited resources in comparison to big biotech companies.
Opportunities: - There is a growing emphasis on preventive healthcare, where early detection and continuous monitoring of health parameters can lead to better health outcomes. This emphasis correlates with the willingness of potentially big markets to spend money on our future biosensors. - According to the data the number of people suffering from kidney problems will increase in years to come. - The current POC devices or laboratory testing does not offer information about the patient's creatinine trends. There is a demand for continuously working biosensors from end-patients as well as from hospitals and other potential customers. In other words, low market saturation. - The requirements of clinical trials in Europe are very high however when completed offer an easier position to enter a variety of markets
Threats: There are big companies (e.g., Abbott Laboratories) or promising start-ups (e.g., Metyos) that have already patented or are developing technology serving the same customer groups. Abbott Laboratories holds patent rights for a device similar to ours. We are currently evaluating the extent of this similarity and determining the feasibility of patenting our device. If necessary, we will make adjustments to ensure patentability. Regulatory challenges can delay product development and market entry.

Appendix 15: MOSCOW Analysis

***Table 7**: MOSCOW analysis.*

Mo	S	Co	W
Must have - The ability to measure creatinine concentration in the ranges of 0.5 mg/dL - 3.0 mg/dL. - The ability to provide reliable, accurate, fast and continuous measurements. - Perfect sealing preventing any leakage (as the flow cell contains a solution of various chemicals). - Cartridge made from non-conducting material/insulating material (as we are using electrochemical detection).	**Should have** - Immobilized enzymes bioassay to extend shelf life and device operation time. - Cost-effective and portable design to make the device more appealing and convenient for non-hospitalized users. - The ability to measure for 7 days a week. - Ability to calculate and show eGFR values. - Safe and durable design. - Mobile application/external display connected to device.	**Could have** - Ability to measure multiple biomarkers (Cystatin C, NGAL) for earlier detection, improved sensitivity and accuracy. - Ability to be used both for kidney malfunction and diabetes mellitus (as the detection principle is relatively the same). - Sustainable materials to reduce CO_2 emissions and reduce environmental burden to minimum. - Ability to be handled by untrained personnel/user.	**Will not have** - Zero environmental burden. - Wireless connection to measuring device (collected samples travel through tubing into the measuring device) -> at least for the current prototype.

Appendix 16: Device Cost Analysis

***Table 8**: Device production costs analysis.*

Device price of laboratory testing prototype			Device price in large-scale manufacturing
Category		**Cost per device**	**Cost**
Flow cell	Plexiglass	€ 2.00	€ 1.60
	Dialysis membrane	€ 1.70	€ 1.36
	Tubing	€ 1.80	€ 1.44
	Glue for plexiglass	€ 0.40	€ 0.32
	Microfluidics pump	*in product replaced by capillary driven flow*	
	Total	**€ 5.90**	**€ 4.72**
Chemicals and enzymes	Creatinine oxidase	€ 159.00	€ 47.70
	Creatine oxidase	€ 16.00	€ 4.80
	Glutaraldehyde	€ 0.30	€ 0.24
	Sarcosine oxidase	€ 12.50	€ 3.75
	Phosphate Buffered Saline (PBS)	€ 0.10	€ 0.08
	Mediator	€ 0.50	€ 0.40
	Cleaning - IPA, MilliQ	€ 0.20	€ 0.16
	Total	**€ 188.60**	**€ 57.13**
External measuring and evaluation hardware	Potentiostat	€ 8.40	€ 6.72
	Batteries	€ 0.50	€ 0.50
	Power supply	€ 5.05	€ 4.04
	Micro connector	€ 0.05	€ 0.04
	Total	**€ 14.00**	**€ 11.30**
Screen printed electrodes *UPCE supplier - accounting for Ink, production, etc.*	€ 2.00		€ 2.00
Others	Cost of labor & assembly	€ 20.00	€ 2.00
	Cartridge 3D printing	€ 1.80	€ 1.80
	Packaging	€ 2.00	€ 1.00
	Delivery and insurance of the shipment	€ 1.25	€ 0.63
	Total	**€ 25.05**	**€ 5.43**
Cost per biosensor / hardware*			
Realistic scenario (expected)		**€ 221.55 / € 235.55**	**€ 67.28 / € 78.58**
Pessimistic scenario (+20% of variable costs)		**€ 265.86 / € 282.66**	**€ 80.73 / € 94.29**
Optimistic scenario (-20% of variable costs)		**€ 177.24 / € 184.44**	**€ 53.82 / € 62.86**

* Hardware is a reading unit that processes and displays all necessary information and data

Given the variable costs for the sensor (+hardware) and market research, the selling price would be set at **€89 per sensor and €109 with hardware**.

All products required for device production are sourced through our established production partners, including Merck, Alchimica, BVT, UPCE, and a few others. The external measuring and evaluation hardware is outsourced, followed by complete in-house hardware assembly. Further details regarding sources and specific mediators used are chosen not to be disclosed.

Appendix 17: Clinical Trials Overview

***Table 9**: Clinical trials overview.*

Trial Purpose	Duration	Participants	Monitoring/Check-ups
Type 2 Diabetes and Weight Loss [32]	6 months	43	4 check-ups
Cancer Patients with Hyperglycaemia [33]	8 weeks	118	Accuracy comparison of the strip test
Type 2 Diabetes Compared to Healthy Subjects [34]	6 months 358 – half of patients over 65 years of age	358 – half of patients over 65 years of age	Assess treatment quality
Paediatric Patients [35]	6 months	62	CGM monitoring & reports
Post Approval Study for Paediatric and Young Adults [35]	6 months	550	CGM monitoring & reports
Discharge Trial [36]	3 months – 3x14 days	100	50 % CGM & 50 % strip tests + CGM – 900 CGM tests, 400 visits, 8 strips a day. 50 readers required.
Insulin-Managed Type 2 Diabetes [37]	8 months	175 – 3 groups (3x, 4x, 6x wearing per 14 days)	CGM monitoring – 754 total CGM
Accuracy Trials [38-40]	Varies	442	CGM & strip tests
Post Approval on Accuracy	3 years	935	CGM & strip tests
Type 2 Diabetes and Medication Adjustment [41]	6 months	300	CGM monitoring

Note: The estimated cost of clinical trials for our future device was calculated based on a preexisting device targeted for continuous glucose monitoring and data available at https://clinicaltrials.gov/

Appendix 18: Clinical Trials Financial Cost Estimates

***Table 10**: Clinical trials financial analysis.*

Cost Component	Units/Hours/Participants	Unit Cost	Total Cost
CGMS	21,687	€ 33.00	€ 715,671.00
Strip Tests	1,534,152	€ 0.50	€ 767,076.00
Readers	1,427	€ 20.00	€ 28,540.00
Visits	6,071	€ 28.00	€ 169,988.00
Intravenous Collections	2,210	€ 14.00	€ 30,940.00
Insurance (10 % of total cost)	-	-	€ 188,343.00
Protocol Costs	9	€ 30,000.00	€ 270,000.00
Data Statistician (3 years)	-	-	€ 153,519.00
Total	**-**	**-**	**€ 2,306,077.00**
Safety margin (+10%)	**-**	**-**	**€ 2,536,684.70**

*Note: Final costs are rounded to €2,540,000 and include delivery fees, duty tolls, and safety margin. The financial cost estimates are based on the clinical trials overview (**Appendix 17**).*

Appendix 19: CE Marking Requirements and Cost Overview

***Table 11**: CE marking requirements overview [42].*

Type	Price	Duration
Risk assessment of consumer product	1,300 €	2 - 4 weeks
Instructions for use	2,500 €	3 - 6 weeks
User manual	4,200 €	4 - 8 weeks
CE marking of medical equipment (class II)	16,900 €	6 - 24 months
Total	**24,900 €**	**-**
Safety margin (+10%)	**27,390 €**	**-**

Appendix 20: Laboratory and Office Rent Overview

***Table 12**: Laboratory and office rent overview.*

Type	Price	Annual cost	Source
120 sqm office	20.5 €/sqm/month	€46,368	[43]
Office services	7.5 €/sqm/month		
14.95 % add-on factor	-		
55 sqm laboratory	50 €/sqm/month*	33,000 €	[44]
Total	**78 €/sqm/month**	**79,368 €**	-
Safety margin (+10%)	**85.8 €/sqm/month**	**87,304.8 €**	-

* Based on the assumption that rental is roughly 50 % cheaper than in Boston [45].

Appendix 21: Marketing Strategy

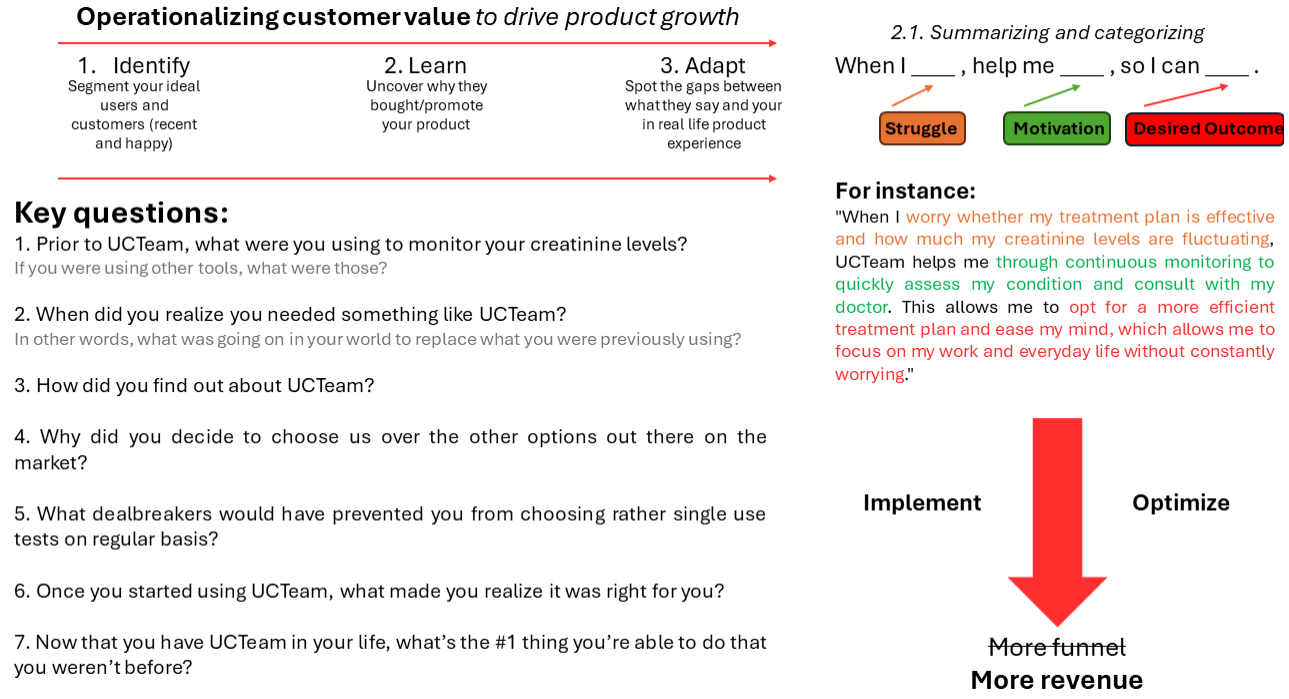


***Figure 12**: Customer-led growth framework overview [18].*

Appendix 22: FMEA

***Table 13**: FMEA initial draft.*

FAILURE MODE AND EFFECTS ANALYSIS

Item:	KidneyGuard UCTeam Biosensor	Responsibility:	J. Cuth	FMEA number:	1
Model:	Prototype	Prepared by:	J. Cuth	Page :	1 of 1
Core Team:	UCTeam			FMEA Date:	05/08/2024 Rev: 1

Item / Function	Potential Failure Mode(s)	Potential Effect(s) of Failure	Sev	Potential Cause(s)/ Mechanism(s) of Failure	Occ	Future Possible Design Controls	Det	RPN	Recommended Action(s)	Responsibility & Target Completion Date	Actions Taken	New Sev	New Occ	New Det	New RPN
Flow cell with screen printed electrodes	Electrode degradation	Inaccurate creatinine measurements, incorrect eGFR values	8	Wear and tear, chemical reactions	6	Impedence measurement to monitor electrode condition	4	192	Develop an electrode lifetime study	R&D team	To be taken based on study results	8	4	3	96
Flow cell with screen printed electrodes	Enzyme activity loss	Reduced sensor sensitivity, inaccurate readings	7	Enzyme degradation over time, improper storage	3	Enzyme activity monitoring through response time analysis	3	63	Conduct stability of enzymes assay	R&D team	To be taken based on study results	7	3	2	42
Flow cell with screen printed electrodes	Chemical reagent depletion	Failure to generate measurable signals	9	Insufficient reagent supply, leakage	2	Reagent level sensor + alerts	2	36	Set up automated alerts for reagent depletion	R&D team, IT department	To be filled in based on implementation	9	2	1	14
Potentiostat	Potentiostat malfunction	Inaccurate voltage measurements, erroneous data	9	Manufacturing defects, assembly issues	3	Verify potentiostat performance before distributing	4	108	Strict assembly procedures, regular training	Engineering department	To be filled in based on implementation	9	5	2	90
Potentiostat	Software errors of potentiostat	Incorrect signal processing, faulty data output	8	Bugs in software, firmware issues	3	Regular software updates, software monitoring with	4	96	Rigorous testing and debugging protocols	IT department	To be filled in based on implementation	8	4	2	64
Tubing	Tubing blockage	Interruption in sample flow, no data	6	Tissue fluid clots, debris buildup	5	Flow sensors monitoring flow rate + alert if low flow detected	5	150	Implement flow sensor with alerts	R&D team, IT department	To be filled in based on implementation	6	3	3	54
Tubing	Tubing disconnection	Data loss, potential exposure to biohazard	8	Mechanical stress, improper connection, overpressure	3	Pressure sensors + alert if sudden pressure drop is detected	4	96	Strengthen and secure connections, add pressure sensor	R&D team, IT department	To be filled in based on implementation	8	4	3	96
Data transmission	Cable damage	Interruption in data transmission	7	Wear and tear, physical damage	4	Visual inspection	4	112	Use more durable cables	Engineering department	To be filled in based on implementation	7	4	3	84
Data transmission	Wireless signal interference	Data loss, transmission delays	6	Device interference, environmental factors	5	Signal strength monitoring	5	150	Implement signal strength monitoring, use frequency-hopping or spread-spectrum technologies to minimize interference	IT department	To be filled in based on implementation	6	4	3	72
Data transmission	Battery depletion	Loss of connectivity, data loss	8	Prolonged use, battery malfunction	3	Battery status indicator and alerts for low battery	2	48	Implement battery status indicators with alerts, use batteries of higher quality	Engineering department	To be filled in based on implementation	8	4	2	64
Data database (AWS)	Server downtime	Inaccessibility of data, potential data loss	9	Maintenance, server crashes	2	Notifications of service status	4	72	Notify users of server issues/downtime	IT department	To be filled in based on implementation	9	2	4	72
Data database (AWS)	Data corruption	Inaccurate or lost data	8	Software bugs	3	Regular data integrity checks and backups	3	72	Implement more rigorous and frequent data integrity checks, set up automated backups, regularly update software	IT department	To be filled in based on implementation	8	3	2	48
Data database (AWS)	Unauthorized access	Data theft, privacy violation	10	Cyber attacks	2	Security breach notifications	2	40	Conduct regular security audits, inform users of security breaches	IT department	To be filled in based on implementation	10	1	2	20
Call centre	Communication failure with healthcare providers	Delayed response to patient needs, potential health risks	8	Network issues, system errors	3	System checks	5	120	Enhance network infrastructure, implement frequent system checks and maintenance	IT department	To be filled in based on implementation	8	2	5	80
Call centre	Inaccurate patient data relay	Incorrect medical advice, potential health risks	9	Human error, data entry mistakes	3	Data reviewed by patient, automated data entry systems, AI to check for given medical advice	4	108	Data reviewed and verified by multiple members/member and AI algorithm	IT department	To be filled in based on implementation	9	2	4	72
Sensor assembly	Incorrect assembly of biosensor	Malfunctioning sensor, inaccurate readings	8	Human error	4	In-process quality checks, automated assembly systems	5	160	Implement in-process checks, implement and improve automated assembly systems	Engineering department, IT department	To be filled in based on implementation	8	2	3	48
Sensor assembly	Incorrect calibration	Inaccurate measurements, unreliable data	9	Calibration equipment failure, procedural errors	3	Calibration process validation, calibration checks	4	108	Upgrade/maintain calibration equipment, stricter calibration process	Manufacturing department	To be filled in based on implementation	9	2	3	54
Packaging and shipping	Damage to package	Device damage during transit, compromised sterility	7	Inadequate packaging materials, improper handling of package	4	Packaging quality inspections	6	168	Use more protective materials, more rigorous packaging	Manufacturing department	To be filled in based on implementation	7	3	6	126
Packaging and shipping	Delays in shipping	Late delivery to customers, unsatisfied customers	5	Logistic issues, carrier delays	5	Real-time tracking of shipments, notifications to customers about delays	5	125	Enhance coordination with logistics providers, inform customers about delays	Logistics team	To be filled in based on implementation	5	4	4	80
Post-market surveillance	Ignoring customer feedback	Unaddressed issues, poor customer satisfaction	6	Inefficient feedback collection process, inadequate follow-up	3	Regular review of customer feedback, automated feedback management systems	4	72	Implement regular reviews, use automated systems	Customer service department	To be filled in based on implementation	6	2	2	24
Post-market surveillance	Delayed reporting of adverse events	Regulatory non-compliance, patient safety risks	9	Inefficient reporting process, lack of awareness	2	Automated reporting system, regular training for staff on reporting procedures	3	54	Implement automated reporting system, provide training for staff	Regulatory affairs department	To be filled in based on implementation	9	1	3	27
Regulatory compliance	Incomplete or incorrect documentation	Regulatory non-compliance, potential fines, prohibition of sale	8	Not keeping up with legal procedures and outdated documents	3	Regular audits of documentation, alerts if new legislative has been approved	4	96	Implement frequent audits, alerts for any regulation changes, keep track of legal requirements	Regulatory affairs department	To be filled in based on implementation	8	2	3	48
Regulatory compliance	Non-compliance with regulatory standards	Legal issues, prohibition of sale	9	Change in regulations, not updating standards	2	Regular regulatory compliance checks	3	54	Implement regular compliance chekcs	Regulatory affairs department	To be filled in based on implementation	9	1	2	18
Software and firmware	Bugs in firmware	Device malfunction, incorrect data processing	8	Inadequate testing, coding errors	3	Comprehensive firmware testing with feedback loops, code debugging	5	120	Increase scope and frequency of testing, use better debugging practices	IT department	To be filled in based on implementation	8	2	4	64
Software and firmware	Failure during software updates	Device downtime, potential data loss	7	Network issues, corrupted update files	3	Rollback mechanisms in case of update failures, data backups	4	84	Implement rollback mechanisms, enhance data backup procedures, review updates before implementation	IT department	To be filled in based on implementation	7	2	3	42
User/customer training and support	Inadequate technical support	User frustration	8	Insufficient support staff, inadequate troubleshooting protocols	3	Regular training for support staff, detailed troubleshooting guideliness and FAQs, assurance of quick response time	4	96	Provide regular training, create comprehensive troubleshooting guidelines and FAQs	Customer service department	To be filled in based on implementation	8	2	4	64

Appendix 23: Financial Analysis

***Table 14**: Financial analysis by phase and year.*

Year	2025-2026	2027-2028	2029-2031	2032-2034	2035-2039	2040
Phase	**R&D**	**Preclinical trials**	**Clinical trials**	**Medical licensing**	**Market launch**	**Market growth**
Fixed costs						
R&D initial costs	€ 150,000.00	€ -	€ -	€ -	€ -	€ -
Patenting [a]	€ 15,000.00	€ -	€ -	€ -	€ -	€ -
Patent maintenance [b]	€ -	€ 8,000.00	€ 12,000.00	€ 12,000.00	€ 20,000.00	€ 4,000.00
Preclinical trials [c]	€ -	€ 132,000.00	€ -	€ -	€ -	€ -
Clinical trials + medical license [d]	€ -	€ -	€ 2,540,000.00	€ -	€ -	€ -
Device risk assessment	€ -	€ -	€ -	€ 1,300.00	€ -	€ -
Instruction manual	€ -	€ -	€ -	€ 2,500.00	€ -	€ -
User manual	€ -	€ -	€ -	€ 4,200.00	€ -	€ -
CE marking class II [e]	€ -	€ -	€ -	€ 16,900.00	€ -	€ -
Laboratory rental	€ -	€ -	€ -	€ -	€ 165,000.00	€ 33,000.00
Office rental	€ -	€ -	€ -	€ -	€ 231,840.00	€ 46,368.00
Salaries [f]	€ 168,000.00	€ 336,000.00	€ 1,008,000.00	€ 1,008,000.00	€ 1,680,000.00	€ 336,000.00
Marketing	€ -	€ 10,000.00	€ 15,000.00	€ 30,000.00	€ 50,000.00	€ 15,000.00
Seminars/ workshops	€ 2,500.00	€ 2,500.00	€ 3,750.00	€ 3,750.00	€ 12,500.00	€ 5,000.00
Fixed costs total	€ 335,500.00	€ 488,500.00	€ 3,578,750.00	€ 1,078,650.00	€ 2,159,340.00	€ 439,368.00
Variable costs						
# of biosensors	-	-	-	-	200,000.00	300,000.00
Variable costs per piece	€ -	€ -	€ -	€ -	€ 78.50	€ 78.50
Variable costs total	€ -	€ -	€ -	€ -	€15,700,000.00	€23,550,000.00
Revenue						
(Revenues price 109 €)	€ -	€ -	€ -	€ -	€21,800,000.00	€32,700,000.00
Funding & grants	€ 335,500.00	€ 150,000.00	€ -	€ -	€ -	€ -
Investors	€ -	€ 338,500.00	€ 3,578,750.00	€ 1,078,650.00	€ -	€ -
Income	€ 335,500.00	€ 488,500.00	€ 3,578,750.00	€ 1,078,650.00	€21,800,000.00	€32,700,000.00
Cash flow	0	0	0	0	€ 3,940,660.00	€ 8,710,632.00
Profit	**-€335,500.00**	**- € 488,500.00**	**- € 3,578,750.00**	**- € 1,078,650.00**	**+ € 2,540,660.00**	**+ € 8,710,632.00**
Cumulative profit	**-€335,500.00**	**- € 824,000.00**	**- € 4,402,750.00**	**- € 5,481,400.00**	**- € 1,540,740.00**	**+ € 7,169,892.00**

[a] patenting costs based on our conducted interview with DTT

[b] patenting maintenance cost based on our interview with DTT

[c] confidential document offer from preclinical studies company

[d] clinical trials overview and cost estimation (**Appendix 17 & 18**)

[e] CE marking requirements (**Appendix 19**)

[f] salary of 1,400 € /month/employee, 5 employees in R&D phase, after R&D phase 10 employees, after preclinical trials 2,800 €/month/employee, future salary increases to position corresponding averages after reaching business profitability

To ensure neutral cash flow during the initial stages, we plan to secure funding through a combination of individual investments, grants, sponsorships, and other financial support. These sources of capital will sustain our operations until we achieve profitability. Once we reach a profitable state, we will begin repaying our investors. According to our projections, we anticipate achieving a positive cumulative profit within 15 years.

***Table 15**: Break even analysis.*

Break even analysis	
Depreciation (total R&D cost 2025-2034/10 years) in €	524140.00
Annual fixed costs without depreciation of R&D costs (2035+) in €	431868.00
Variable costs in €	78.50
Price in €	109.00
Break-even point (annual)	**31344.52**

For our break-even analysis, we need to sell a **total of 31345 devices** to cover all costs and reach break-even. This translates to selling approximately **2612 devices per month annually**. Key details: **Production cost per device is €78.50**, and the sale price **per device is €109.00**. All initial costs are accounted for and amortized over the 10-year fixed period, ensuring that each device sold contributes towards covering the cumulative expenses.

Appendix 24: Comprehensive Financial Analysis Expense Breakdown (2025 – 2040)

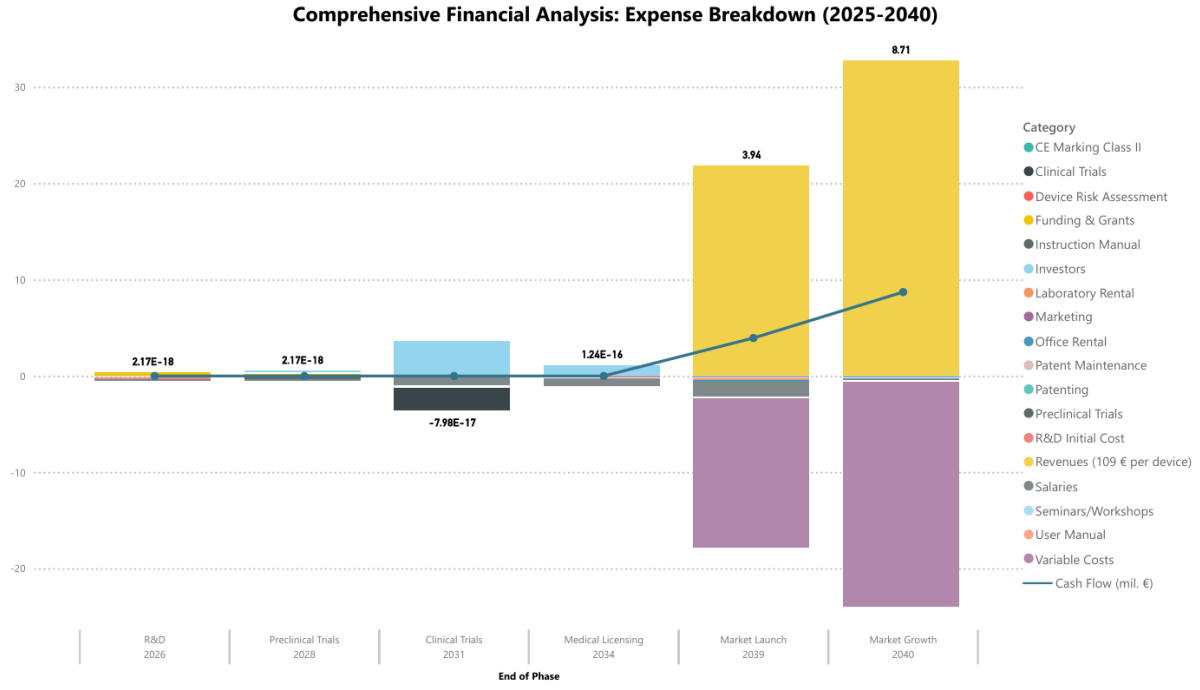


***Figure 13**: Comprehensive financial analysis breakdown.*

List of Abbreviations

3D	Three-Dimensional
ACR	Albumin Creatinine Ratio
ADC	Analogue-Digital Converter
AKI	Acute Kidney Injury
ARF	Acute Renal Failure
AWS	Amazon Web Service
BSA	Bovine Serum Albumine
CA	Chronoamperometry
CE	Counter Electrode
CE marking	Conformité Européene marking
CEO	Chief Executive Officer
CGM	Continuous Glucose Monitor
CKD	Chronic Kidney Disease
CLEA	Cross-Linked Enzyme Aggregates
CSc.	Candidate of Science
CT	Computed Tomography
CTA	Clinical Trial Authorization
CZ	The Czech Republic
DAC	Digital-to-Analog Converter
DC	Direct Current
doc.	Associate Professor
DTT	Department of Technology Transfer
e.g.	exempli gratia
ECEIM	The European College of Equine Internal Medicine
EDS	Energy-Dispersive X-ray Spectroscopy
eGFR	Estimated Glomerular Filtration Rate
EMA	European Medicines Agency
ER	Emergency Room
ERA congress	European Renal Association congress
EU	European Union
FGF23	Fibroblast Growth Factor-23
FIR	Finite Impulse Response
FMEA	Failure Mode and Effects Analysis
FZU	Czech Academy of Life Sciences
GLP	Good Laboratory Practices
GNP	Graphene Nanoplates
GP	General Practitioner
GUI	Graphical User Interface
HPLC	High Performance Liquid Chromatography
ICU	Intensive Care Unit
IDMS	Isotope Dilution Mass Spectrometry
Ing.	Inženýr (Czech) / Engineer (English)
IPA	Isopropyl Alcohol
ISF	Interstitial Fluid
ISO	International Organization for Standardization

KT	Kidney Transplant
LC	Liquid Chromatography
LLC	Limited Liability Company
MDR	Medical Device Regulation 2017/745
MIPs	Molecularly Imprinted Polymers
MOSCOW analysis	Must- Should- Could- Will not- have analysis
MRI	Magnetic Resonance Imaging
MS	Mass Spectrometry
MUDr.	Doctor of Medicine
MVDr.	Doctor of Veterinary Medicine
NGAL	Neutrophil Gelatinase-Associated Lipocalin
PBS	Phosphate Buffered Saline
PCB	Printed Circuit Board
Ph.D.	Doctor of Philosophy
PMMA	Polymethyl Methacrylate
POC	Point of Care
R&D	Research and Development
REF	Reference Electrode
RNDr.	Doctor of Natural Sciences
SAM	Serviceable Addressable Market
SDMA	Symmetric dimethylarginine
SOM	Serviceable Obtainable Market
SPE	Screen Printed Electrode
SPCE	Screen Printed Carbon Electrode
sqm	square meter
SWOT analysis	Strengths, Weaknesses, Opportunities and Threats analysis
TAM	Total Addressable Market
TRL	Technology Readiness Level
TV	Television
UART	Universal Asynchronous Reciever/Transmitter
UCT	University of Chemistry and Technology, Prague
UPCE	University of Pardubice
US	United States
UX/UI	User Experience/User Interface
WE	Working Electrode

List of Symbols

A	Electric Current [A]; 1 mA = 10^{-3} A; 1 µA = 10^{-6} A
C	Carbon (Chemical Element)
M	Concentration/Molarity [M = mol L^{-1}]; 1 µM = 10^{-6} M
V	Voltage [V]; 1 mV = 10^{-3} V
H_2O	Water (Molecular Formula)
H_2O_2	Hydrogen Peroxide (Molecular Formula)
O_2	Oxygen (Molecular Formula)
Re(III)	Rhenium(III) (+3 oxidation state of Rhenium)
Re(IV)	Rhenium(IV) (+4 oxidation state of Rhenium)
ReO_2	Rhenium Oxide

List of Tables

List of Figures

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